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PREFACE

Difficulties of maintaining longevity and productivity of stone fruit trees have been the motivation for three workshops to study problems associated with declining trees. In the first Stone Fruit Decline Workshop, which was held at Michigan State University in 1982, scientists from several disciplines and many fruit-growing regions met to discuss cultural and physical factors and biological agents that are responsible for decline in productivity and premature death. That meeting served to underscore the many factors that can and do result in debilitation of fruit trees. The papers presented and the discussion that followed pointed out the urgent need for interdisciplinary and inter-regional cooperation to address effectively the complex and diverse nature of stone fruit tree decline in North America.

The Second Stone Fruit Decline Workshop was held at the USDA/ARS Appalachian Fruit Research Station, Kearneysville, West Virginia in 1984. This workshop focused primarily upon biological agents and their interaction with each other and the host trees that they infect. Practices and processes leading to infection and the host responses in microbial invasion were studied. Again, the diversity of organisms involved was evident from the wide range of topics that were discussed. Participants agreed that a single cause or even several factors could not explain the progress of decline in stone fruits in all areas of North America. Even single agents that were clearly important in decline problems in a region or locality appeared to be interacting with other agents or affected trees that were under stress.

The next logical step for consideration of stone fruit decline was to identify and study stress factors that render fruit trees susceptible to pathogens, pests, or physical factors in the environment. Therefore, factors in the orchard environment that impose stress upon stone fruit trees became the focus for presentations at the Third Stone Fruit Decline Workshop, which met at the Ramada Inn in Clemson, South Carolina, October 28-29, 1986. This Proceedings contains most of the papers that were presented at this workshop.

I wish to thank all of the participants in this workshop for their helpful attitudes and patience in preparing their manuscripts for publication.

Eldon Zehr, editor
Clemson, SC

CONTENTS

STRATEGIES AND IMPLEMENTATIONS OF PEACH GROWTH

J.G. Williamson and D.C. Coston[1]

Controlling vigor of tree fruits has been a major concern of pomologists for centuries. Achieving a proper balance between vegetative and reproductive growth is necessary to enhance earliness and maximize yields during the life of the orchard. Without acceptable dwarfing rootstocks, a suitable means for controlling vigor of peach has not been determined. Summer pruning and various growth regulators have offered some suppression of vigor, yet these practices can be costly and often have adverse side effects.

A system was developed in Australia (Chalmers et al.,1981) to reduce vigor of peach without adversely affecting yields by practicing regulated deficit irrigation during stages 1 and 2 of fruit growth followed by full irrigation rates during stage 3. A semiarid climate coupled with a shallow top soil and an underlying clay pan allowed for more precise control of tree growth via irrigation rates than could be achieved in the humid Southeastern United States.

Confinement of the peach root system to a small volume of soil should allow for more precise control of the tree's water status during periods of maximum vegetative growth.

This paper reports the results of 2 studies conducted to improve our knowledge of controlling vigor of peach. The objectives were as follows: (1) To observe the relationships among shoot growth, fruit growth and root growth of peach, and (2) To determine the effects of root restriction on vegetative and reproductive growth of peach trees.

MATERIALS AND METHODS

Experiment 1. Twenty root observation boxes were established in the field at the South Carolina Agricultural Experiment Station in Clemson, South Carolina. The boxes were 80 cm high, 55 cm wide, and 30 cm thick and contained approximately 0.1 m^3 of Cecil sandy loam soil. The boxes were constructed of 0.5 in. plywood and covered with 2 in. sheets of Styrofoam. The fronts of the boxes were constructed of bilayer safety glass inserted at a slight angle from top to bottom to encourage root development at the soil-glass interface. The glass surfaces were covered with sheets of Styrofoam wrapped in foil and held in place with adhesive strips. Eight of the root boxes contained tensiometers at 15 cm, 30 cm and 45 cm depths. Four of the root boxes contained thermisters positioned against the inside surface of the glass at 15 cm and 30 cm depths. Shoot growth, fruit growth and root growth were measured daily for 8 of the 20 boxes from May 24 until September 3 in 1985 and from May 2 until July 16 in 1986. Weekly measurements for shoot growth, fruit growth and unsuberized root length were taken on all 20 boxes during the 1985 and 1986 growing seasons. Shoot and fruit growth were measured using previously described methods (Haun and Coston, 1983). Daily root growth was determined by tracing all root growth within a 945 cm^2 area in 1985 and a 400 cm^2 area in 1986 on acetate sheets affixed to the glass fronts. Root tracings were measured in the laboratory with a Zeiss Interactive Digital Analysis System (Carl Zeiss, Thornwood, NY) to determine total root growth, number of growing roots, and growth per root for each 24-hr. period. Weekly unsuberized root length was determined by the line intersect method (Head, 1966) and taken to be an indication of root activity. Unsuberized root length was measured at less frequent intervals during the fall and winter of 1985-1986.

Experiment 2. A high-density peach orchard was established at the Clemson Agricultural Experiment Station during the summer of 1984. Prior to planting, the orchard was limed, subsoiled, disced, rototilled and fumigated with methyl bromide. Own-rooted 'Redhaven' peach trees were planted at an in-row spacing of 1 meter with 2 meters between rows. Each plot contained 15 trees (3 rows of 5 trees). Data were collected from the three center trees in the middle row of each plot.

Five treatments were initiated at the time of planting: 1. Control - trees set in small holes just large enough to accommodate the root system. 2. Auger planting hole - trees set in 45 cm deep hole dug with a 20-cm auger under wet soil conditions. 3. Raised bed - top soil pulled from between rows into rows before planting to form beds approximately 1.0 m wide by 20 cm high. 4. Narrow herbicide strip (NHS) - trees planted like controls with 0.5-m-wide herbicide strips. Herbicide strips for all other treatments were maintained at a width of 1.0 m. 5. Fabric - Trees were planted in trenches 90 cm wide by 30 cm deep which were lined with a synthetic polyester impregnated with an acrylic latex. Overhead and trickle irrigation were used during the remainder of the 1984 growing season to minimize transplant shock. During the 1985 growing season all plots were trickle irrigated identically. In the spring of 1986, 2 irrigation treatments were imposed on each planting treatment. The high irrigation treatment replaced the estimated daily evapotranspiration (ET) based on class A evaporation pan readings during all stages of fruit growth, while the low irrigation treatment replaced 12.5% of the estimated ET during stages 1 and 2 of fruit growth and 100% of the estimated ET during stage 3.

Morning (6:00 - 8:00) and afternoon (1:30 - 3:30) leaf water potential readings were taken with a Scholander pressure bomb during stage 2 of fruit growth and again during stage 3. The 2 irrigation treatments were reimposed during late summer of 1986 following a period of relatively

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frequent rains and AM and PM leaf water potentials were taken at weekly intervals from September 8 until October 1.

The experiment was conducted as a randomized complete block with 7 replications.

RESULTS

Experiment 1. Flushes of shoot growth were followed by periods of increased unsuberized root length in the 1985 and 1986 growing seasons (Fig. 1). A detailed examination of the 1985 growing season revealed 2 flushes of shoot growth, each followed by a period of rapid root growth (Fig. 2). Shoot growth and root growth were inversely correlated (r=-0.620). Total unsuberized root length was similar for fruited and nonfruited trees with the exception of a period of 2-4 weeks during the end of stage 3 of fruit growth and immediately following harvest (Fig. 3). Total unsuberized root length was greatest for fruited and nonfruited trees during late summer after vegetative growth had ceased but before leaf abscission occurred.

Experiment 2. In 1984, vegetative growth was reduced by the auger planting treatment as evidenced by reduced plant height, trunk caliper and number of lateral shoots per tree (Table 1). Although the auger planting treatment resulted in smaller trees throughout the 1985 growing season, this effect was no longer evident by the end of the 1986 growing season. However, in 1985, the NHS and fabric treatments resulted in less vegetative growth than other treatments. This pattern was continued in the 1986 growing season (Table 2). Flower bud density was substantially increased by the fabric treatment and reduced by the NHS treatment (Table 1.) Internode lengths were shorter for the auger, NHS and fabric planting treatments than for the control or raised bed treatments. However, flower bud density was also influenced by the number of flowers per node. The high flower bud density of the fabric treatment was accompanied by a significant increase in flowers per node. Fruit weight, number of fruit per tree, and yield per tree were similar for the fabric and the control treatments even though the fabric trees were much smaller.

Morning leaf water potentials taken during stage 2 of fruit development were lower (more negative) for those treatments receiving low irrigation rates regardless of planting treatment (Table 3). Afternoon leaf water potential readings taken during stage 2 were lower for the fabric (100% ET replacement) than for the control (100% ET replacement) readings during stage 3 even though all treatments were receiving 100% ET replacement at this time. Afternoon leaf water potential readings taken during stage 3 were similar with neither the NHS nor the fabric treatments differing significantly from the control.

Late season AM leaf water potential readings taken at weekly intervals showed no difference for NHS or fabric treatments when compared to the control of either irrigation rate (Table 4). However the PM water potential readings for the fabric (100% ET replacement) were lower than the control (100% ET replacement) at all measuring dates (Table 5). The fabric (12.5% ET replacement) had lower leaf water potential values than the control (12.5% ET replacement) at 1 of the 4 measuring dates.

DISCUSSION

Experiment 1. The strong relationship between shoot growth and root growth observed in this study suggests a high degree of integration of growth within peach trees. Competition within the plant for photosynthate is probably a major factor causing the flushes of root and shoot growth. Shoots, being nearer to the source, can presumably compete more efficiently than roots. However the amount of shoot growth that can occur without subsequent root growth is limited by the root system's ability to supply the shoot with essential growth components (i.e. water, mineral salts, and hormones). Therefore root growth must occur if shoot growth is to be sustained. The observed flushes of root and shoot growth in this study are in agreement with work done with citrus (Bevington and Castle, 1985). However, this study had sufficient replication to add statistical validity to the results, something commonly lacking in other root research. The influence of cropping on root growth observed in this study is in general agreement with work on apple (Head, 1969). However, the reduction in root growth from cropping was temporary with typical root activity resuming by 3 or 4 weeks following harvest.

Experiment 2. The reduction in vigor achieved from the NHS and fabric treatments persisted for 2 growing seasons. Although the NHS treatment produced smaller trees, these trees had low flower bud densities and less fruit per tree than control or the fabric treatments. Conversely, the fabric treatment produced trees of small size with considerably higher flower bud density than other treatments and had adequate fruit set. Planting treatments had a greater effect on vigor than irrigation rates. Afternoon leaf water potential readings taken at the end of the 1986 growing season suggest that the trees in the fabric treatment reached a greater degree of stress than the control trees. This is probably due to the inability of the root system to exploit large volumes of soil for water during periods of high transpiration.

The mid-day leaf water potential values for the fabric (100% ET replacement) treatment and fabric (12.5% ET replacement) treatment were lower than those values observed for the control (100% ET replacement). The higher mid-day water deficit of these treatments could in turn limit photosynthesis and other growth related processes. Another possibility is altered nutrient and/or hormone transport from the root system.

CONCLUSION

Most plant species have a characteristic root:shoot ratio. Therefore, a reduction in

root growth or root function should result in a corresponding reduction in top growth.

The maintenance of a strong root:shoot ratio in peach is supported by the high degree of integration of root and shoot growth observed in experiment 1.

Root restriction appears to be an effective method of reducing peach tree vigor, probably due to a reduction in root growth or function. Further experimentation is needed to determine the most appropriate method of root restriction and its long term effects on tree performance.

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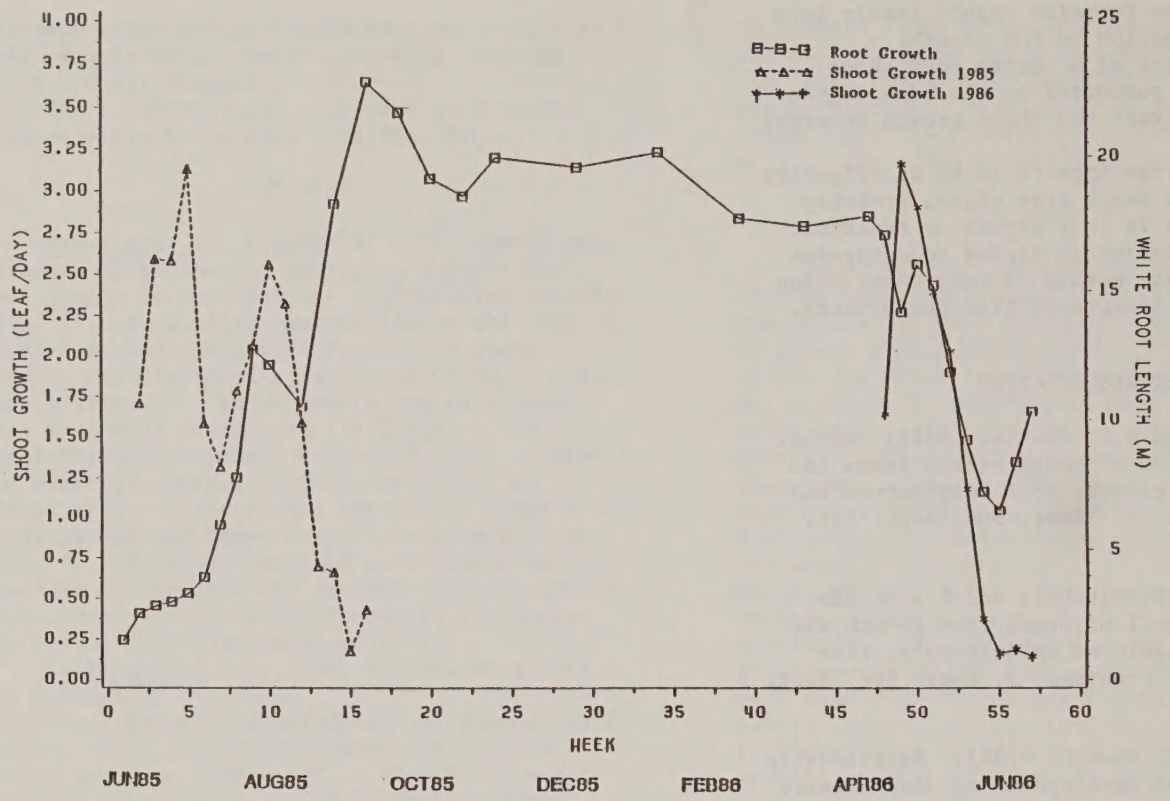

Figure 1. The influence of shoot growth rate on total white root length of 'Redhaven' peach trees.

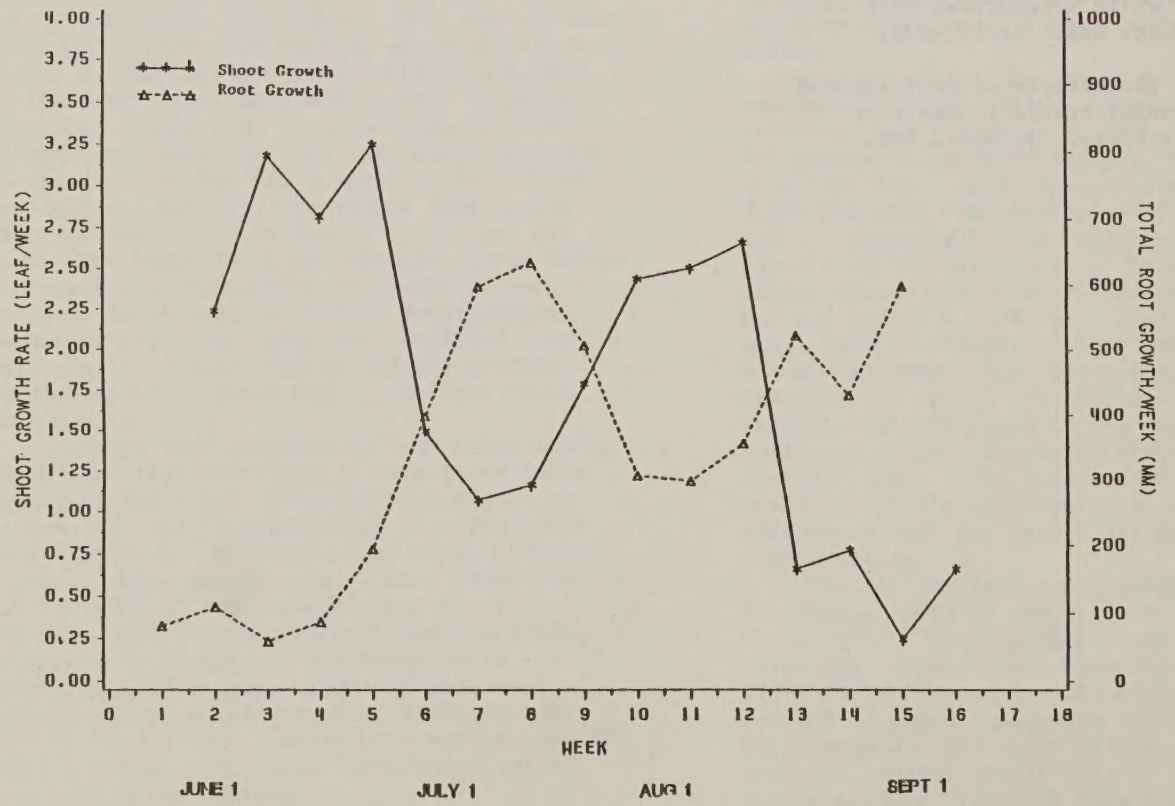

Figure 2. Mean values for weekly shoot growth rates and weekly root growth rates of nonfruited peach trees.

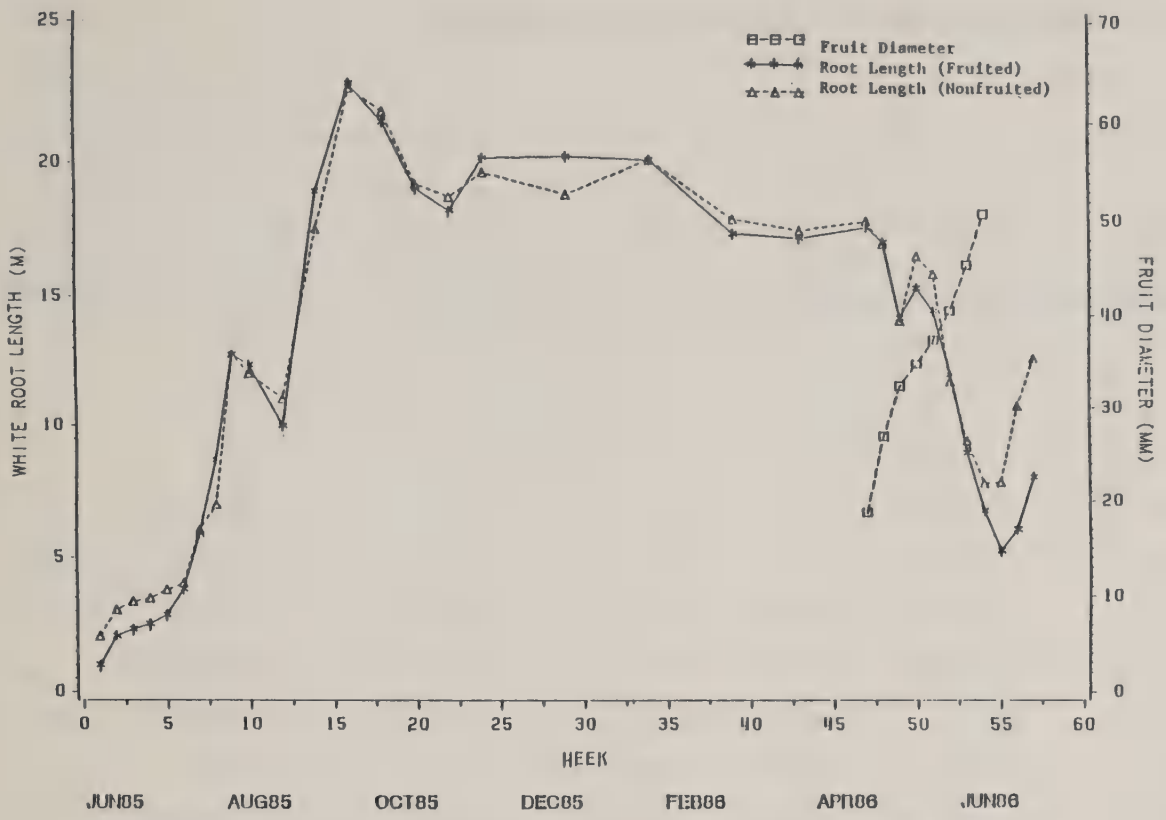

Figure 3. The influence of fruit growth on total white root length of 'Redhaven' peach trees.

Table 1. Effect of planting treatments on growth and development of 'Redhaven' peach trees in 1984 and 1985.

	1984			1985					
Treatment	Plant height (cm)	Trunk diameter (cm)	Number of laterals/plant	Plant height (cm)	Trunk diameter (cm)	Canopy width (cm)	Internode length (cm)	Flowers/cm shoot length	Flowers per node
Control	45.5[z]	1.0a	4.6a	216.6a	4.0	154.7a	1.9a	0.17bc	0.30b
Auger	32.4b	0.8b	1.4b	191.7b	3.5	143.6b	1.8bc	0.14c	0.25b
Beds	45.8a	1.2a	5.3a	210.0a	3.7	154.1a	1.9ab	0.18b	0.34b
NHS	45.8a	1.1a	4.3a	193.4b	3.2	125.6c	1.7c	0.15bc	0.27b
Fabric	49.8a	1.1a	6.0a	183.4b	3.1	120.2c	1.8bc	0.26a	0.45a

[z]Letters denote mean separation in columns by LSD test, 5% level.

Table 2. Effect of planting treatments and irrigation rates on vegetative and reproductive growth of 'Redhaven' peach in 1986.

Planting treatment	Percent ET replaced	Tree height (cm)	Canopy width (cm)	Trunk diameter (cm)	Number of fruit/tree	Fruit Weight (g/fruit)
Control	100	288.7a[z]	181.3ab	4.7a	45.4ab	65.5abc
Control	12.5	283.7ab	183.9a	4.7a	57.6a	69.2abc
Auger	100	276.9abc	181.7ab	4.5ab	35.6bc	72.8a
Auger	12.5	255.3cd	173.3abc	4.2bc	36.5bc	68.1abc
Bed	100	280.0ab	194.0a	4.5abc	52.6a	66.3abc
Bed	12.5	280.8ab	181.2ab	4.7a	44.6ab	68.5abc
NHS	100	262.3bcd	183.0ab	4.1bc	27.0c	66.3abc
NHS	12.5	253.8d	162.3bcd	4.0c	24.3c	71.2ab
Fabric	100	218.0e	158.9cd	3.6d	57.3a	61.9c
Fabric	12.5	205.1e	146.8d	3.4d	43.7ab	62.8c

[z]Letters denote mean sepaaration in columns by LSD test, 5% level.

Table 3. Effect of planting treatments and irrigation rates on leaf water potential of 'Redhaven' trees during stage 2 and stage 3 of fruit growth in 1986.

		Stage 2 of fruit growth		Stage 3 of fruit growth	
Planting treatment	Percent ER replaced	AM (kPa)	PM (kPa)	AM (kPa)	PM (kPa)
Control	100	-3.4b[z]	-15.8bc	-2.7a	-16.0ab
Control	12.5	-8.1a	-19.7ab	-4.1b	-16.3ab
NHS	100	-2.9b	-14.2c	-2.5a	-14.6b
NHS	12.5	-7.4a	-18.7abc	-3.2a	-15.0b
Fabric	100	-4.8b	-23.4a	-3.0a	-19.3a
Fabric	12.5	-7.7a	-17.2bc	-3.2a	-16.7ab

[z]Letters denote mean separation columns by LSD test, 5% level.

Table 4. Effect of planting treatments and irrigation rates or late season AM leaf water potential values in 1986.

Planting treatment	Percent ET replaced	Date: September 8 (kPa)	September 15 (kPa)	September 24 (kPa)	October 1 (kPa)
Control	100	-2.5ab	-3.0a	-2.1a	-2.5ab
Control	12.5	-2.7a	-2.7a	-2.1a	-2.8ab
NHS	100	-2.3ab	-3.2a	-2.7a	-2.1b
NHS	12.5	-2.6a	-3.1a	-1.7a	-2.6ab
Fabric	100	-2.5ab	-3.3a	-2.4a	-2.2b
Fabric	12.5	-2.1b	-3.3a	-2.8a	-3.3a

[z]Letters denote mean separation in columns by LSD test, 5% level.

Table 5. Effect of planting treatments and irrigation rates on late season PM leaf water potential in 1986.

Planting treatment	Percent ET replaced	Date: September 8 (kPa)	September 15 (kPa)	September 24 (kPa)	October 1 (kPa)
Control	100	-15.3b	-12.5c	-14.9ab	-16.4cd
Control	12.5	-17.5ab	-15.0abc	-15.7b	-18.2abc
NHS	100	-16.1b	-13.1bc	-14.8b	-15.8d
NHS	12.5	-15.2b	-13.6bc	-14.2b	-17.6bcd
Fabric	100	-20.7a	-15.8ab	-18.2a	-19.1ab
Fabric	12.5	-15.1b	-17.1a	-17.8a	-20.0a

[z]Letters denote mean separation in columns by LSD test, 5% level.

SOIL PH AND NUTRITION AFFECT PEACH TREE YIELD AND LONGEVITY

George A. Cummings[1]

In a recent review article Yadava and Doud (1982) thoroughly reviewed the literature including effects of soil pH and nutrition on peach tree longevity. They cited numerous studies where nutrition or foliar levels were associated with some attribute of tree performance. However, only in regard to nitrogen nutrition and soil pH did they so much as imply a cause-effect relationship between nutrition and peach tree longevity. Our work in North Carolina supports conclusions of their review. In controlled field experiments with various soil amendments of N, P, K, B, Ca, Mg, Zn, Cu, Mn, and lime as well as tillage variables and foliar sprays, only three practices have been identified that are associated with peach tree short life. They are:

- Low soil pH
- Nitrogen - too little (25 years), too much (one year)
- Low magnesium supply

The effect of low Mg was easy to identify. In that experiment, with three levels of K and Mg, soil pH was maintained with calcitic lime in a soil that was inherently low in Mg. Trees without a Mg amendment were so deficient in Mg that defoliation of basal leaves occurred b mid-summer. Severity of the deficiency increased as K application rates increased. Obviously this malady could be easily prevented by maintaining soil pH with dolomitic lime.

NITROGEN

Peach trees grown in a humid climate are similar to any other non-leguminous crop in their need for supplemental N. However, the amount required to supplement the N in the soil may, and does, vary greatly from year to year. Numerous factors may account for this. The first is utilization of N applied the previous year. Regardless of the form of N applied, ammonia or nitrate, it is necessary for the ions to move into the rooting area. Nitrogen deficiency is often observed in the late summer even though trees have been supplied with adequate N. In 1986, due to prolonged drought in mid-summer, many orchards in N. C. appeared to be N-starved in late summer. The second factor relates to fruit load. A heavy crop may account for nearly as much N removed as does tree growth and foliage. Finally, we have observed many times that orchards established in fields which have been heavily N fertilized the prior year may require very little N during the first year. Undoubtedly there is some residual N in the soil.

Starting in 1954 we at North Carolina State University have carried out numerous nitrogen rate and source experiments. In every experiment prior to 1984 tree growth and yield increased as N rate was increased. In addition, longevity either was not affected, or trees lived longer with the higher N rates. We found in a 20-year-old orchard that had been fertilized with 0.4, 0.8, or 1.6 pounds of N per tree after the fourth year that the highest N rate had the best survival, though not significantly higher than the 0.8-pound rate. However, survival at 0.8 or 1.6 pounds was significantly higher than at the 0.4-pound rate. Other nitrogen experiments supported the hypothesis of increased longevity with increased N supply. However, there is another side of the nitrogen story.

In two large experiments established in 1978, one consisting of 512 trees (a two to the seventh factorial), results for the first six years relative to growth, yield, and survival were similar to results of previous experiments. Variables in the first experiment were: subsoiling, B, Cu, Zn, ordinary or super phosphate, pH 5.8 or 6.2, N source of ammonia or nitrate, and N rates of 0.64 or 0.96 lbs of N per mature tree. In the other experiment variables were: subsoiling, P rates, K rates, foliar B sprays, soil applied Cu, Zn, and B, N rate of 0.48 or 0.64 lb per tree, and time of fertilizer application - two-thirds March, one-third after harvest or one-third March, two-thirds after harvest. In the first experiment significant increases from subsoiling, high pH, and N rate were noted by 1981 in regard to tree growth and yield. Survival was high in all treatments until 1983 as shown in Table 1. However, cold injury on December 24, 1983 resulted in severe trunk injury in all treatments. The low temperature occurred after a very mild autumn and late tree defoliation. Trees were vigorous regardless of treatment, but especially so in the high N and subsoil treatments. Only in the case of high soil pH was survival enhanced and losses in the high N treatment were the most severe. Losses resulting from high N were probably also enhanced by the lack of a fruit crop during the 1983 season. Similar results from high N occurred in the second experiment even though N rates were lower.

Table 1. Effect of subsoiling, pH, and nitrogen rate on tree survival 1978-85. (512 trees)

	Number of trees					
	Subsoiling		pH		N-rate	
Date	No	Yes	5.8	6.2	0.64	0.96
6-83	246	249	244	251	245	250
8-84	202	210	199	213	217	195
5-85	87	81	61	107	111	57

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In summary one might interpret from experimental results with N that longevity appears to be associated with tree vigor and the time during the dormant period when injury occurs. With the exception of extremely vigorous growth in late fall, which should always be avoided, our data show that usually trees with relatively high nitrogen have survived better than low-N-status trees. Another factor to be considered is that damage to high-N trees is probably much greater if the low temperature occurs early compared to later in the dormant season. We feel confident that many more trees are lost in N. C. from low compared to high-N rates. However, vigorous growth late in the growing season should always be avoided.

In the two experiments established in 1978, and in many other nutritional experiments, no other nutritional factors have been related to tree longevity. In two different experiments phosphorus application prior to establishment and during the first three years has tended to increase survival, but there was no difference in survival after the trees reached maturity.

SOIL pH

In numerous experiments with peaches in N. C. and throughout the Southeast, I am not aware of any deleterious effects of lime applied to acid soil. Almost always pH maintained above 6.0 compared to 5.5 has been associated with increased tree growth, fruit yield, fruit size, and tree longevity. Occasionally, when all of the above measurements are made, lime has not proved superior every year, but the overall favorable effects are quite convincing. Possibly the first necessity is to understand the nature of soil acidity.

Why are soils acid? The first and primary reason is simply that base cations, Ca, Mg, and K have been leached from the surface soil as water passes through the soil profile. These ions are replaced with hydrogen and ultimately aluminum. When pH is measured one is simply measuring the hydrogen concentration in the soil. However, the real culprit probably is not hydrogen, but aluminum. In an acid soil, below pH 5.0, the cation exchange complex may be 70 to 90 percent saturated with aluminum. When soils are limed, Ca and Mg replace the aluminum and the exchange complex is base-saturated. In N. C. Sandhill soils at pH 6.5, 70 to 90 percent of the exchange complex is Ca or Mg with almost no aluminum.

The second reason soils are acid results from the use of acid-forming N fertilizers. When ammonia nitrogen (the dominant form of N in commercial fertilizers) is applied to soil the end result is nitrate and hydrogen:

ammonium + oxygen ——> nitrate + water + hydrogen

In general each pound of N from ammonia sources lowers pH as much as 1.8 pounds of lime raises it. The pH depression from ammonium sulphate and ammoniated phosphates are approximately 3-fold greater than from ammonium nitrate. The depression of soil pH can be avoided if basic sources of N such as calcium, potassium, or sodium nitrate are used, but it is normally cheaper to use ammonia sources and neutralize the resulting acidity by lime application.

Two other losses of bases from soils are crop removal and erosion. With fruit crops, calcium and magnesium content of fruit is low. Losses from these sources are usually minor with fruit crops.

What happens when lime is applied to soil? Nothing, until the limestone reacts with soil acids. Limestone is hard, durable, and not soluble in water. (Some of our oldest buildings in this country are made of limestone.) Its purity is measured on the basis of pure calcium carbonate = 100. If good dolomitic limestone is used the calcium carbonate equivalent may be well over 100. However, unless it is finely ground it is worthless as a means of improving soil pH. In N. C. in order for limestone to be sold as a liming material it must conform to the following:

- Ninety percent must pass through U.S. standard 20-mesh screen.
- For dolomitic lime 35% must pass through a 100-mesh screen; for calcitic lime 25% must do so.
- Must have 6% magnesium to qualify as dolomite; and
- The label must state clearly amount of material needed to equal 90% calcium carbonate equivalent

Assuming that one applies good agricultural lime and mixes it with the soil, it reacts, the pH is raised and it 1) decreases soluble Al, 2) decreases soil Mn, 3) decreases K leaching, 4) raises soil Mg, 5) raises soil Ca, 6) improves soil tilth, 7) alters soil microflora, and 8) alters rooting area.

Each of these effects probably is helpful for peach tree performance. The most important point listed is aluminum. It has been shown for a number of non-leguminous crops that aluminum is deleterious to root growth. Similar results have been observed at Byron from the work of Edwards and Horton (1977) with peaches. Aluminum is not a problem if soil pH is above about 5.7.

Manganese in soil behaves similarly to aluminum. However, it is an essential element and is absolutely required for tree growth. Various problems occur with many crops when Mn levels are too high. In N.C. I usually do not need a soil test to estimate soil pH if I have a foliar analysis. As a general guide, if foliar Mn is 80 ppm or under, soil pH is probably 6.0 or above, if 80 to 150 ppm, pH is probably 5.5 to 6.0, and if above 150 ppm, pH is probably below 5.5. This may vary in peaches grown in other areas because some soils are extremely high in manganese (many Piedmont soils in the Southeast). I doubt if foliar levels of 100-250 ppm hurt the tree, but it would probably be better off at lower Mn levels.

It has been shown that K losses from leaching are markedly reduced when acid soils are limed. Although not listed, phosphorus availability is increased as pH is increased up to about 6.5.

The next two points, foliar levels of Ca and Mg, are important in the case of Mg because it is often deficient in southeastern soils. However, I believe that the relationship may be an artifact - that the favorable effect often attributed to high foliar Ca levels may be related to some other factor related to soil pH. Why, without proof, should we associate a favorable result from liming to increased foliar Ca when a myriad of other changes also occur with a change in soil pH?

We have done almost no work with soil microflora or improvement in the soil tilth. However, we have noted a greater proliferation of roots below the normal plow sole after pH modification of that area.

What favorable effects of increased pH are supported by experimental data? We started two experiments in the fall of 1961. One was a field with a history of tree short life, and that experiment was terminated in 1966. The other was established in a field that had been fallow for several years, was replanted again in 1972, and terminated in September 1986.

The first experiment was only one of five experiments in a large field. Severe tree loss occurred after the second year and this also occurred with our lime treatments. Lime was applied in the winter of 1961-62 to establish pH of 4.9, 5.4, and 6.0, and another application of 1000 lb per acre was applied to all plots in the winter of 1963-64. Soil pH at termination of the experiment after the 1965 season was 5.2, 5.4, and 5.7 respectively. Even though losses were high the effect of lime was evident (Table 2). Tree growth, yield and survival as well as fruit size (not shown) were improved with increasing soil pH. However, since losses were large in all treatments we paid little attention, at that time, to these differences.

Table 2. Effect of soil pH on peach yield, tree growth and longevity (1962-66).

Original soil pH	Trunk Dia. 5 yr	Yield Kg/tree	% Survival	Final soil pH	Foliar Ca - %
4.9	2.94	39.3	42	5.2	0.84
5.4	3.54	54.0	58	5.5	0.95
6.0	3.52	53.7	67	5.7	1.18

The effect of lime in the second experiment that was established in 1961-62 was evident by the third year with greater tree loss in the unlimed soil. Loss of trees continued until 1970 in all treatments but was much less at the two higher pH levels. However, losses regardless of pH were heavy in 1971, all trees were pulled, the soil was fumigated that fall and replanted in the spring of 1972.

We terminated this experiment in September 1986, and longevity is recorded in Table 3. Yields in 1981, the last year detailed harvest records were kept, as pH was increased were: 123, 213, and 340 bushels per acre; weight per peach 145, 147, and 153 grams; peaches per tree 265, 313, and 454; and trunk diameter 8.9, 10.1, and 12.5 cm, respectively. Calcium and magnesium levels also increased as pH increased and at the highest pH reached 1.95% Ca and 0.47% Mg. The favorable effect on yield per acre was enhanced because of higher yield per tree as well as more living trees per acre. These data show the long term effect of soil pH and these effects have been widening since the early years of the experiment. However, we often notice the effect on longevity and tree growth starting the first year and the effect on fruit yield and size the first harvest year.

We also carried out two experiments starting in 1965 and the same results from increased pH on fruit size occurred (Table 4). However, in both 1965 experiments pH was amended to the desired pH

Table 3. Effect of soil pH on peach tree longevity (25 years).

pH	% Live trees Sep. 85	% Live trees Apr 86	Annual lime lb/acre
5.2	17	6	380
5.7	27	19	585
6.2	54	43	800

prior to establishment and again in the winter of 1967-68, but no more lime was applied and soil pH in 1975 was 5.1 and 5.3, respectively. Even in these experiments the favorable effects of lime during the establishment years were evident even though all treatments were very acidic by 1975. Total bushels per acre as pH increased in experiment one was 946 and 1466 per acre and in experiment two, 1133 and 1348 bushels per acre, respectively. The increased soil pH enhanced longevity in the first but not in the second experiment.

Table 4. Effect of soil pH on peach size (1967-72).

Soil pH	Wt per peach (grams) Year 1967	1968	1969	1970	1971	1972
5.5	135	143	150	114	108	104
6.2	135	149	168	120	115	126

The experiment previously described in regard to nitrogen, a two to the eighth factorial, was designed to answer several questions. The reason for the pH variable was obvious, the phosphate variable furnished two levels of Ca, and the Zn, Cu, and B were included to determine the effect of a soil pH micronutrient interaction. Foliar Ca levels were increased by use of high Ca phosphate and calcium nitrate, but these variables did not effect longevity. Micronutrients also did not effect tree performance. As shown in Table 1 only soil pH appeared to have an effect, although minor, on tree longevity. Thus it is obvious that altering soil pH in acid soils cannot guarantee survival. Yet, regardless of year or other factors we have in general noted every attribute of neutralizing soil acidity is a plus for tree performance.

Finally, none of our tests, including many not listed here, and with the exception of soil pH, have really had a great effect of tree longevity. Maintaining soil pH above 6.0 will enhance tree survival but is only one of many practices needed to optimize tree longevity. We have paid great attention to general fertility in fruit crops. Fertility is important, and a change in fertilizer can greatly affect tree performance. However, if one takes any sufficiency range published for essential nutrients and pick a value within that range as ideal, I would probably find no fault with the choice. For example, we have observed good tree performance with N at 2.2% as well as 3.2% and with K at 1.4% as well as 2.6% (we have experimental results to support all figures). We do not have that much range with soil pH - at least not in N.C. Results from all our pH variables indicate almost certain failure at pH 5.5 or below, curtailment of yield, tree growth, fruit size and increased tree death from 5.5-6.0, and optimum results only when pH is maintained at 6.0 or above.

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ROOT DISTRIBUTION OF NON-IRRIGATED AND TRICKLE-IRRIGATED PEACH TREES

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INTRODUCTION

Trickle irrigation systems are becoming very popular in the southeastern US, especially for fruit and vegetable crops. Properly designed and operated systems minimize water and energy requirements, yet provide ample water to the crop. Since optimum system operation requires measurement or estimation of soil moisture tension within the root zone, it is necessary to have reliable information on the distribution of crop roots. If, for example, irrigation scheduling is to be accomplished with tensiometers, the most effective tensiometer placement (depth, radius from tree, and distance from emitter) depends on knowledge of root distribution. Also, if mathematical models are used to estimate soil moisture, allocation of water withdrawal among various portions of the root zone would be important and would be related to the distribution of roots.

The purpose of this project was to characterize, for two common soil types in South Carolina, the dependency of peach root density on depth, radius from tree, orientation angle from tree (in-row or cross-row), and radius from emitter.

REVIEW OF RELATED LITERATURE

The root growth of most fruit plants is limited by species and age of plant, soil type, restricting soil layers, depth to water table, nutrients, aeration, cultural practices, and competition from neighboring plants (Rogers and Head, 1966, 1969). For most of the studies conducted on peaches, the maximum depth of root penetration has been limited by a restricting soil layer. Even without such a layer, however, almost all of the roots have been found in the top 60 cm.

Havis (1938) dug a trench radially from several trees and counted the number of roots extending from the trench wall. He found that 56-65% of the total number of roots were in the top 30 cm of soil, and 85-92% were in the top 60 cm. Maximum root penetration was 90 cm for three of his trees, but one tree in a deep soil had a few roots as deep as 200 cm. This is consistent with Olsson (1977), who found 55% of the total root length in the top 30 cm, 80% in the top 60 cm, and almost no roots below 125 cm. Cockroft and Wallbrink (1966) observed that the highest concentration of root length occurred in the top 30 cm of soil for their trees, but that the highest weight concentration was in the 30- to 60-cm depth interval. They found few roots below 90 cm and concluded that the massive clay B horizon was responsible for the shallow rooting. Examinations of the clay showed that the fine roots were confined to cleavage planes and large pores, which suggested a problem of mechanical impedence.

Some studies have shown that there is a reduction in root concentration very near the surface (Proebsting, 1943). This is probably due to high temperatures, which have been shown to prevent growth of peach roots in sand culture (Nightingale, 1935), and also to periodic drying below the wilting point.

According to Rogers and Head (1966), the roots of most fruit plants tend to first fill a cylindrical volume with a diameter of 1.5 to 3 times the branch spread. Later, in commercial orchards, this cylinder becomes more nearly a square or rectangle, delimited by root competition from adjacent trees. Data reported by Havis (1938) shows little effect of radius on the number of small feeder roots in a 10-year-old orchard planted on a 6.1-m spacing.

Although peach root production is enhanced by moist soil conditions (Richards and Cockroft, 1975), almost no information is available on the effect of trickle irrigation emitters on peach root distributions in humid regions. In semi-arid Australia, Willoughby and Cockroft (1974) and Taylor (1974) reported that peach roots did not thrive (and some roots were even killed) directly under trickle emitters, probably due to poor aeration in the saturated zone. Instead, roots tended to accumulate at the periphery of the wetted zone. This was also observed for tomatoes (Goldberg et al., 1976) and wheat (Silverbush et al., 1979). Edwards et al. (1982) did not find evidence for this phenomenon in peach roots (except when NH_4NO_3 was added to the irrigation water), but they did observe a higher average root concentration within a 50-cm radius of the emitter. This was also observed by Chesness and Couvillion (1986), who found the root concentration within 50 cm of the emitter to be 2.7 times that in a non-irrigated zone.

Finally, if one is ever able to characterize the root distribution of peach trees, the more difficult problem of relating moisture withdrawal to root distribution remains. Although it is commonly believed that water is absorbed through unsuberized young roots, suberized roots can account for 60-96% of the total water absorbed by trees and other woody perennials (Kramer and Bullock, 1966). A more complete review of this topic is given by Atkinson (1980).

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EXPERIMENTAL EQUIPMENT AND PROCEDURES

Two sites were chosen for this study. The first was a Hiwassee sandy loam in the Piedmont region at Clemson, SC. The other site was a Norfolk loamy sand in the upper Coastal Plain at Edgefield, SC.

The Hiwassee sandy loam is a well-drained soil in broad upland areas on long side slopes. Typically, the surface layer is dusky-red sandy loam about 15 cm deep. It is dark-red sandy clay loam between 15 and 38 cm, dark-red clay between 38 and 90 cm, and dark-red clay loam below 90 cm.

The average annual rainfall at this site is 138 cm. A winter maximum normally occurs in January and February; a smaller summer maximum normally occurs in July and August. The fall is dry and the spring relatively dry. Evaporation normally exceeds precipitation in the summer, which may include short and extended periods of drought. The average maximum temperature (over the entire year) is 22.5°C; the average minimum is 9.2°C.

The trees were 'Redglobe'/'Lovell' planted in 1976 on a 6.1-m x 6.1-m spacing. Each row lay in the center of a 3.0-m herbicide strip. One emitter per tree was installed in 1979; each emitter was placed in the center of the herbicide strip about 1.5 m from the tree trunk.

Norfolk loamy sand is a deep, well drained, nearly level soil found on broad upland areas of the Coastal Plain. Typically, the surface layer is grayish brown loamy sand about 18 cm thick. The subsurface layer is light yellowish brown loamy sand 20 cm thick. The subsoil extends to a depth of 180 cm or more. It is yellowish brown sandy loam between 38 and 50 cm and yellowish brown sandy clay loam between 50 and 100 cm. It is red sandy clay loam that has yellowish brown mottles between 100 and 115 cm and red clay loam that has yellowish red and reddish yellow mottles between 115 and 180 cm.

The average annual rainfall at the Norfolk site is 118 cm, and is distributed in a manner similar to that described for the Hiwassee site. The average maximum temperature is 23.3°C, and the average minimum is 9.2°C.

The trees were 'Babygold-8'/'Lovell' planted in January 1980 at a spacing of 6.1 m x 6.1 m. Each row lay in the center of a 3.0-m herbicide strip. A trickle emitter was installed near each tree in 1980 and a second emitter was added in 1981. The trickle line was buried 20 cm deep and 1.2 m from the tree row; the two emitters were 1.2 m on either side of the tree.

Both sites were sampled in February and March of 1985. At each site, two trickle irrigated trees and two non-irrigated trees were sampled. A hydraulic press was used to extract 7.5-cm diameter cores from depths to 150 cm. From each sampling hole at the Hiwassee site, cores were divided into samples with depth intervals of 0-25, 25-50, 50-100, and 100-150 cm. At the Norfolk site, the depth intervals were 0-25, 25-50, 50-75, 75-100, and 100-150 cm. At each tree, four holes were sampled along the tree row line at radii of 37, 112, 187, and 262 cm from the tree. A second set of four holes were sampled at the same radii from the tree, but along a line perpendicular to the tree row (crossing the grassed traffic lane). For the irrigated trees, in addition to the eight holes described above, three additional holes were sampled at radii of 0, 30, and 60 cm from the emitter (but at a constant radius from the tree). The samples from each site were stored at 4°C until they could be processed.

A simple but effective machine was constructed to separate the roots from the soil. Wire mesh (1/16-inch) was wrapped around a wooden frame to form a cylinder 35 cm in diameter and 60 cm long. The ends of the cylinder were closed. The cylinder was partially submerged in a water bath, with the axis of the cylinder lying horizontally and just above the water surface. A 0.06-kW electric motor was used to rotate the cylinder. A soil/root sample was put in the cylinder and the cylinder was closed and mounted. The cylinder, partially submerged, was rotated at 40 rpm. This was the maximum speed at which the soil would fall from the top of the cylinder as it rotated, rather than becoming "plastered" to the screen by centrifugal force. After the water flowing out of the cylinder was clear (2-3 minutes), the cylinder was opened and the roots were removed from the screen with tweezers. Grass roots were discarded, and the peach roots were dried at 95°C for 48 hours. Feeder roots (<=2 mm diameter) were weighed separately from structural roots (>2 mm).

RESULTS AND DISCUSSION

Analysis of variance was conducted on feeder root weights (<=2 mm diameter) to evaluate the root concentration relative to depth, radius from tree, angle from tree, presence or absence of irrigation, and radius from emitter (Table 1).

Depth. Most of the feeder roots were found near the surface for both soil types and for both irrigated and non-irrigated roots (Figs. 1-4, Table 2). High-clay subsoil layers restrict root growth to the top 125 cm, except for a few cleavage planes. A majority of the roots were found in the top 25 cm. For the deeper Norfolk soil; however, a higher proportion of roots was found in the 50-100 cm interval than for the Hiwassee soil.

The extent of shallow rooting suggests that tillage operations could result in serious damage to the root system, and perhaps should be avoided. Irrigation strategies should seek to maintain optimum moisture conditions in the top 25-50 cm. As long as the water content in this zone is properly maintained, the value of any further irrigation is questionable. The possible

Table 1. Effects of various factors on peach feeder root density.

Test	Hiwassee		Norfolk	
	Tree	Emitter	Tree	Emitter
(a) depth	**	**	**	**
(b) radius	**	0	**	0
(c) depth x radius	**	0	**	0
(d) angle	0		0	
(e) radius x angle	0		0	
(f) depth x angle	0		0	
(g) depth x radius x angle	0		0	
(h) irrigation status	0		0	
(i) emitter vs. tree	0		0	

** = significant at 0.05 level
* = significant at 0.10 level
0 = not significant at 0.10 level

Table 2. Distribution of peach feeder roots with depth.

Depth (cm)	Hiwassee		Norfolk		Depth (cm)
	Tree	Emitter	Tree	Emitter	
0-25	64%	65%	61%	62%	0-25
25-50	24	26	17	6	25-50
			10	10	50-75
50-100	9	5			
			7	19	75-100
			5	3	100-125
100-150	3	4			
					125-150
	100%	100%	100%	100%	

exception to this is trickle irrigation in a deep soil, where deeper root growth may be promoted in the trickle zone (Atkinson, 1980).

Radius. For both soils, feeder root density was shown to decrease with increasing radius from the tree (Figs. 1 and 3; Table 1, line b). Radius from the emitter, however, was a different story. For the Norfolk soil, the data suggests that root density is lower immediately adjacent to the emitter than it is at the edge of the wetted zone (Fig. 4), although radius from the emitter was not statistically significant at the 0.10 level for the entire wetted zone (Table 1). Sandy soil adjacent to a buried emitter would probably be saturated during irrigation, and thus would not be conducive to root growth. A lower root density near the emitter was not observed in the Hiwassee data, however. This is because the soil around the emitter probably does not become saturated. The emitter lies on top of the ground, allowing the water to spread out before infiltrating the surface rather than being pumped into one location. Also, the tendency of heavier soils to trap air would make saturation less likely.

Angle. No difference in root density was detected between root density samples taken in line with the row and those taken in a line perpendicular to the tree row crossing into the grassed traffic lane (Table 1, line d). Also, no interactions were detected between angle and radius or depth. In other words, angle was not found to affect the root distribution with radius or depth (Table 1, lines e-g).

Irrigation status. The feeder root density of non-irrigated roots was unaffected by the irrigation status of the tree (Table 1, line h). There is little evidence that irrigation of one part of the root zone would retard or promote root growth in the rest of the root zone, especially if the non-irrigated roots are receiving ample rainfall to support root growth, as is probably the normal case in the humid regions (Atkinson, 1980).

Emitter vs. tree. When the feeder root density near the emitter was compared to the root density of non-irrigated roots (at the same radius from the tree as the emitter), the differences were not found to be significant (Table 1, line i). This is inconsistent with the findings of Edwards et al. (1982) and Chesness and Couvillion (1986). Perhaps differences would have been more apparent had the sampling been done in August, when the roots are active, rather than in March, when they have been dormant for five months. The maximum difference in new root production between irrigated and non-irrigated roots should occur in the summer, when the amount of soil water is a major limiting factor to root growth in non-irrigated soil (Richards and Cockroft, 1975; Willoughby and Cockroft, 1974). During the fall and early spring, however, much of this difference may be negated by the resumption of new root production in the non-irrigated zone and/or the shedding of roots in the trickle zone (Cockroft and Olsson, 1972; Head, 1973). Another factor could be the method used to obtain root density data. If a zone of maximum root development exists at the edge of the wetted area, it is possible that such a zone would be by-passed by the discrete point sampling method used in this study. Perhaps a trench study would be more appropriate for studying root proliferation around an emitter.

SUMMARY AND CONCLUSIONS

Field sampling in two South Carolina peach orchards showed that most of the roots are found in the top 25 cm, which suggests that irrigation strategies should be directed toward the top 25-50 cm. Also, tillage operations could result in serious damage to peach root systems, and should perhaps be avoided. The proliferation of roots

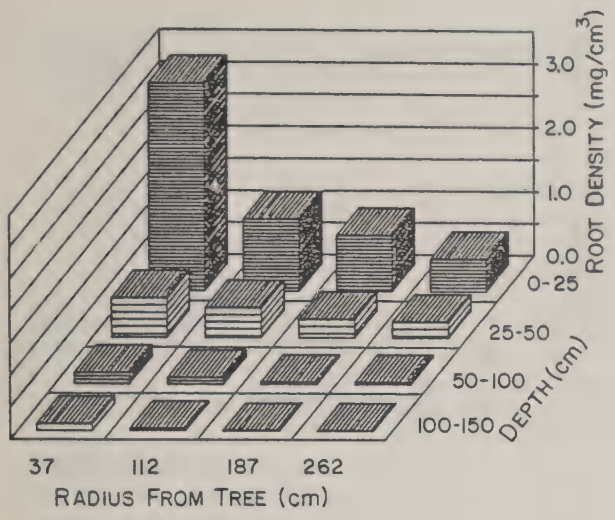

Figure 1. Mean root density (measured in mg roots per cm^3 soil) vs. depth and radius from tree in Hiwassee soil.

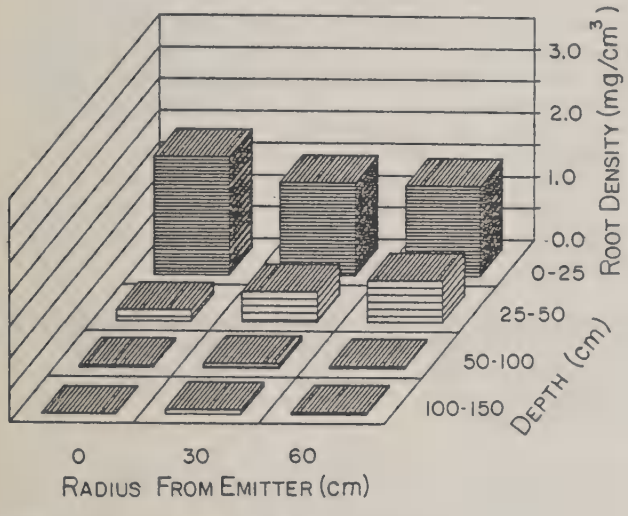

Figure 2. Mean root density vs. depth and radius from emitter in Hiwassee soil.

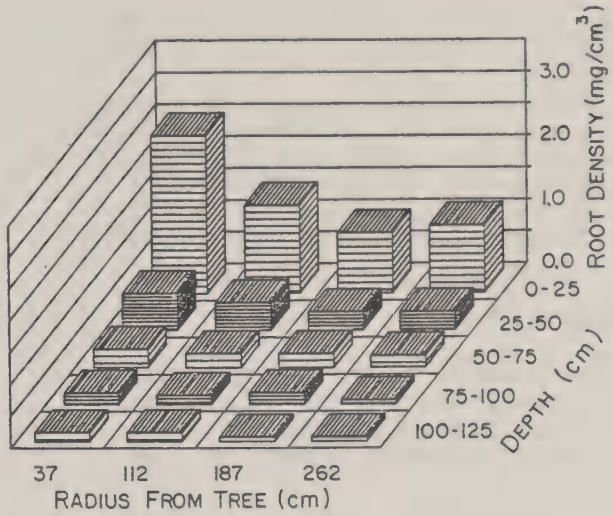

Figure 3. Mean root density (measured in mg roots per cm^3 soil) vs. depth and radius from tree in Norfolk soil.

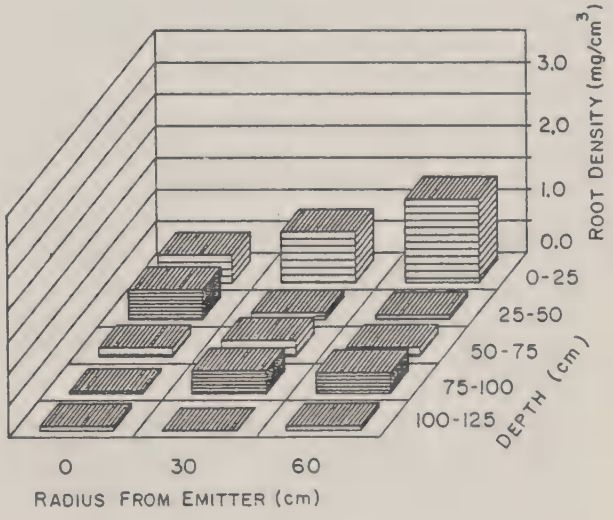

Figure 4. Mean root density vs. depth and radius from emitter in Norfold soil.

in the surface layers decreased with increasing radius from the tree.

The effect of trickle irrigation on the root system was uncertain. No differences in root density were detected between irrigated and non-irrigated roots. For buried emitters in the Norfolk loamy sand, the data suggested fewer roots immediately adjacent to the emitters.

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PEACH TREE AND NEMATODE RESPONSES TO VARIOUS SOIL TREATMENTS UNDER TWO IRRIGATION REGIMES

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ABSTRACT

Tree growth, fruit production and population dynamics of root knot nematode, Meloidogyne spp., pin nematode, Paratylenchus hamatus, and stubby-root nematode, Paratrichodorus minor, were evaluated over a six-year period in a new planting of peach, variety Loadel, on Lovell rootstock. The treatments were: 1) preplant soil fumigation with methyl bromide at 450 kg/ha + mycorrhizae at planting, 2) root-knot nematode + Dactylella oviparasitica (S isolate), 3) root-knot nematode + soil from an established peach orchard, and 4) root knot nematode only. These four treatments were evaluated under a normal and a high irrigation regime. The methyl bromide treatment showed the greatest positive impact on tree growth, fruit production, and nematode control. There was no positive impact from the use of D. oviparasitica; rather, there appeared to be a negative impact in comparison to the root-knot-nematode only treatment. The use of a high irrigation regime greatly counteracted the effects of root-knot nematode. In fact, there was an increase in overall crop value in the high irrigation regime versus the normal irrigation regime for all treatments. Presently, this increase in crop value would more than compensate for the increase in irrigation costs in the high irrigation regime in the San Joaquin Valley.

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ESTIMATED PEACH TREE DEATH IN SOUTH CAROLINA SINCE 1979 - CAUSES AND ECONOMIC IMPACT.

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The peach industry is important to the agriculture economy of South Carolina. It is generally the fifth largest crop in the state. Several diseases severely impact the life of an orchard, reducing current and future production as well as profitability as an agriculture enterprise.

Priorities should be set so as to have maximum impact with scarce resources. Two attributes are important in setting research priorities for impact. They are, which diseases are having maximum negative effect on production and profitability and where will the application of resources yield usable information reducing the effect of disease most rapidly.

No research resources have been expended to regularly document the extent of severity of diseases that result in tree death in South Carolina. References in the literature regarding South Carolina losses represent the opinion of professional peach workers, do not segregate causes, and were not done in regular fashion.

The Extension peach pathology program maintains a cadre of individuals who are competent in the diagnosis of disease, both public and private. The existence of a body of technology which reduces or manages the risk associated with the operation of a peach enterprise is necessary both for these individuals and to maintain the competitive edge of the peach enterprise.

The objective of this program is to assist with research priority setting, both with respect to other commodity programs as well as within the peach research effort.

METHODS:

Annually the observations and opinions of trained professional peach workers, both public and private, are amassed to create the estimates below. Information used varies with the year and programs in effect. Sources included the records and observations of public and private scouts, plant pest regulatory personnel, county and area agents, pesticide industry personnel, and those of the author. The usual pattern is for the experienced observer to drive every 5th row middle, developing an individual estimate for each orchard.

RESULTS:

An average of 161,405 (3.9%) trees were lost annually ranging from a low of 75,000 to a high of 284,280 (Table 1). For the seven year period from 1980 to 1986 this represents a total loss in potential income of $93,422,815. Peach tree short life represents $55,926,722 of this loss or almost eight million dollars per year. Oak root rot represents $30,790,869 or almost 4.4 million dollars annually. Crown rot results in the least identified damage or impact, $4,527,612 or 0.65 million dollars per year.

Regional differences are apparent from the observations. Peach tree short life is the most frequent cause of death in the ridge area of the state, followed by the coastal plains and the piedmont (Table 2). Oak root rot is most severe in the piedmont followed by the coastal plains and the ridge (Table 2). Crown rot is the least in the ridge with about equal severity in the piedmont and coastal plains. All other problems, even when lumped together are small in relative severity and impact (Tables 1 and 2).

DISCUSSION:

The number of trees in a region impacts on the number of trees killed due to methodology. The piedmont acreage is now contracting due to poor financial position of some producers. Nearly 350,000 trees were abandoned in 1985-1986. It is anticipated that possibly a similar number may be abandoned in 1986-1987. The tendency is for old unproductive orchards or young orchards with poor tree population density to be removed. These orchards are usually associated with poor management, and the remaining orchards with better management. This may be impacting percent dead trees (Table 1).

The relative high level of oak root rot in the piedmont is probably due to several interacting factors. Ring nematodes are predisposition agents to peach tree short life. Ring nematodes reproduce slower in piedmont soils, resulting in reduced incidence of short life and hence longer living trees.

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Such trees are probably at greater risk from oak root rot due to longer exposure time and hence greater opportunity for becoming infected with oak root rot.

Many observers have noted an apparent association of oak root rot with the more poorly drained sites on a state-wide basis. Some have hypothesized that "wet feet" or crown rot or possibly both may be a significant interacting factor in the amount of oak root rot observed.

Strains of oak root rot are present, as indicated by carpophores with and without annulus.

Phytophthora cinnamomi is constantly associated with crown rot in South Carolina, and crown rot is associated with poorly drained sites. It may be more significant than the figures indicated as noted above.

Table 1. Estimated Peach Tree Losses in S. C., 1980 - 1986.

Region and Counties Considered	No. Orchards Observed	No. Trees Observed	No. Trees In Region[1]	Percent Trees Dead[2]
Piedmont:	1980: 300	300,000	1,600,000	1.5
Oconee,	1981: 220	220,000	1,600,000	2.7
Pickens,	1982: 254	420,000	1,600,000	2.2
Greenville,	1983: 326	326,000	1,850,000	6.4
Spartanburg,	1984: 350	350,000	1,850,000	3.5
Cherokee,	1985: 110	110,000	1,750,000	3.0
York	1986 100	100,000	1,400,000	3.2
Ridge:	1980: 400	350,000	1,300,000	3.4
Aiken,	1981: 700	703,000	1,430,000	2.1
Edgefield,	1982: 162	163,000	1,500,000	3.6
Saluda,	1983 258	258,000	1,713,000	8.8
Lexington	1984 300	300,000	1,713,000	9.5
	1985: 107	107,000	1,700,000	9.1
	1986: 120	120,000	1,700,000	4.8
Coastal Plains:				
Barnwell,	1980: 50	5,000	340,000	2.0
Allendale,	1981: 30	3,000	414,000	1.8
Hampton,	1982: 30	3,000	420,000	1.5
Orangeburg,	1983: 165	165,600	440,000	3.4
Sumter	1984: 130	130,000	460,000	1.9
Chesterfield	1985: 25	2,500	500,000	2.4
	1986: 35	3,500	550,000	2.1
Total 1980:	750	700,000	3,240,000	2.3
Total 1981:	950	953,000	3,444,000	2.2
Total 1982:	446	586,000	3,520,000	2.4
Total 1983:	749	750,000	4,002,000	6.2
Total 1984:	780	780,000	4,002,000	4.9
Total 1985:	242	242,000	3,950,000	5.6
Total 1986	255	255,000	3,650,000	3.8

[1]The last tree census was in 1982. The numbers reported include estimated trees planted, abandoned or pushed up since 82.

[2]Does not include trees pushed up the previous winter.

Table 2. The estimated causes of tree death in South Carolina by year of death and region

Causes[1]	Percent Tree Death by Year						
	1980	1981	1982	1983	1984	1985	1986
Piedmont:							
Short life	37	15	59	7	14	16	26
oak root rot	36	83	39	91	70	64	54
crown rot	24	1	Tr	Tr	14	16	17
rosette, phony, yellows, stem pitting	Tr	Tr	Tr	1	Tr	Tr	Tr
other (undiagnosed)	3	1	2	Tr	3	4	3
Ridge:							
short life	60	93	96	77	88	83	87
oak root rot	20	6	3	20	10	14	11
crown rot	10	0.3	Tr	2	1	2	1
rosette, phony, yellows, stem pitting	Tr	Tr	Tr	0.5	0.7	Tr	Tr
other (undiagnosed)	9	0.5	0.6	0.1	Tr	Tr	Tr
crown gall	Tr	0.2	Tr	Tr	Tr	Tr	Tr
Coastal Plains:							
short life	60	50	30	90	23	31	26
oak root rot	20	37	40	1	22	28	49
rosette	1	1	Tr	Tr	Tr	Tr	Tr
crown rot	10	3	Tr	4	28	30	14
phony yellows, stem pitting, crown gall	3	3	3	3	3	3	3
other (undiagnosed)	Tr	Tr	Tr	Tr	Tr	Tr	Tr
	6	5	26	0.8	24	8	7%

[1]The cause of death was determined by individuals who made field observations.

NOTE: Tr = Trace

EFFECT OF FALL AND WINTER PRUNING AS RELATED TO CULTIVAR IN PEACH TREE SURVIVAL

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INTRODUCTION

Many investigations have shown that pruning peach trees before they are fully dormant is harmful (Chandler and Daniell, 1976; Daniell, 1973 and 1978; Dowler and Petersen, 1966; Nesmith and Dowler, 1975, 1976; Prince and Horton, 1972; Ritchie and Clayton, 1981; Weaver et al., 1974, 1976), but many peach growers who have large acreages find it impractical to delay pruning until January or February. It is generally accepted that pruning peach trees before they are fully dormant increases stress and may lead to early tree mortality (Carter, 1976, 1978). Previously, workers have looked at time of pruning in association with other stress-inducing factors, such as ring nematode infestation, and have concluded that pruning should be delayed well into the winter (Nesmith and Dowler, 1975, 1976).

Some cultural practices seem to mitigate or enhance the injurious effects of fall pruning. Dowler and Petersen (1966) found that inoculation of pruning wounds with Pseudomonas syringae pv. syringae after pruning in October resulted in tree mortality, while noninoculation resulted in less injury. Little injury followed inoculation of February-pruned trees. Ritchie and Clayton (1981) found that less mortality resulted if November-pruned trees were growing in D-D- and DBCP-fumigated soil compared with nontreated, nematode-infested soil. In 1974, Weaver et al. reported severe tree loss following November pruning in a replanted orchard site but none in soil that was not an orchard site previously (1974). Earlier studies have not compared the responses of different cultivars to pruning at different times, and some studies have been carried out in soils infested with the ring nematode, which by itself may be responsible for early mortality (Lownsbery et al., 1973; Nyczepir et al., 1983; Weaver et al., 1974; Wehunt et al., 1980). The study reported here was designed to evaluate the effect of time of pruning on tree survival as related to chilling requirements of peach cultivars. Four cultivars with different chill hour requirements were tested. We wished to determine, if possible, early, nonlethal pruning times for the 4 cultivars, represented as either a chronological date or a value of chill unit accumulation.

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MATERIALS AND METHODS

An orchard was established in Lakeland fine sand on a short-life site where peach trees had been grown previously and had died from peach tree short life (PTSL), at Clemson University's Sandhill Research and Education Center near Elgin, South Carolina. To control ring nematode infestation, 3.7-m-wide strips of soil were preplant fumigated with 1,3-dichloropropene (Telone II, The Dow Chemical Company, Midland, MI) at a rate of 336 liters/ha in November, 1977. In February, 1978, 312 peach trees on 'Lovell' rootstock, consisting of 4 cultivars having different chilling requirements -- 'Junegold', 'Camden', 'Redglobe', and 'Redhaven' (650, 750, 850, and 950 chill hours, respectively), were planted in a randomized complete block design. During February, 1979, the trees were pruned lightly, and any trees of a cultivar which exhibited atypical phenotypic characteristics or which had not survived, were replaced. Orchard soil was postplant fumigated with Nematocide solution 17.1 containing 2.05 kg 1,2-dibromo-3-chloropropane (DBCP)/liter (AMVAC Chemical Corporation, Los Angeles, CA) in November, 1981, at 28 liters/ha (EPA experimental use Permit No. 5481-EUP-1), by shank injection in a 2-m band within 1 m of the trunk on each side of the tree row.

Beginning in the fall of 1980 and each fall and winter for 4 years, trees were randomly selected from each cultivar, excluding border rows, and pruned according to standard grower practice (Brittain and Miller, 1978). Pruning was performed on five dates in late fall and winter during 1980 and on a biweekly basis during each of the next 3 years, with 4-6 trees of each cultivar pruned on each date.

Throughout each fall-winter pruning season, chilling units were recorded according to the description of the Utah chill unit model (Richardson et al., 1974). Fall pruning was initiated after positive chill unit acquisition had begun (late October) and continued until the end of February. During April, May, and June, pruned trees were evaluated visually for cold injury to the cambium in the trunk and lower scaffold limbs and for other symptoms of PTSL. Obvious indications of PTSL were failure of trees to leaf out in the spring or sudden wilting after leaf growth had begun. Suspect trees were further examined by cutting into the tree to cambial depth at several sites and noting vascular cambium coloration and wood odor. Browning of the vascular cambium and presence of a "sour-sap" odor were confirming indicators of PTSL (Chandler et al., 1964; Prince, 1966). Dead trees were removed from the orchard before the subsequent pruning season.

RESULTS AND DISCUSSION

Death from cold injury was high for all cultivars pruned in October, November, and December, and diminished to an average of 8% among trees

pruned in January (Table 1). Death was reduced to less than 1% by pruning in February. All cultivars responded similarly to pruning over the 4-year period, but the pattern of tree loss in 'Redglobe' differed from the other three cultivars. Relatively few 'Redglobe' trees died in 1981, 1982, and 1984, but many died in 1983 (Table 2). The pattern of tree loss for the other cultivars was the opposite. This pattern of mortality might have been related to the pattern of chill unit accumulation (Fig. 1), which in 1982-83 was characterized by unusually warm weather in November and December. Peach growers evidently would endanger cultivars of low or high chilling requirement by pruning them in the fall.

These data support many previous reports that pruning peach trees in autumn or early winter is harmful to their survival (Chandler and Daniell, 1976; Daniell, 1973, 1978; Nesmith and Dowler, 1975, 1976; Prince and Horton, 1972; Ritchie and Clayton, 1981; Weaver et al., 1974, 1976). Peach growers are urged not to prune before January 1 (Ritchie and Clayton, 1981) or even February 1 (Daniell, 1978; Spivey and McGlohon, 1973), but the need to delay pruning often conflicts with other farming practices, especially for farms having large acreages. To accommodate the needs of farmers, perhaps it is advisable to suggest that least valuable orchards (old trees, less productive trees, or orchards decimated by tree loss) be pruned first. Pruning time as related to tree age probably needs further investigation, and other effects of pruning time (fruit yield and tree growth) should be explored.

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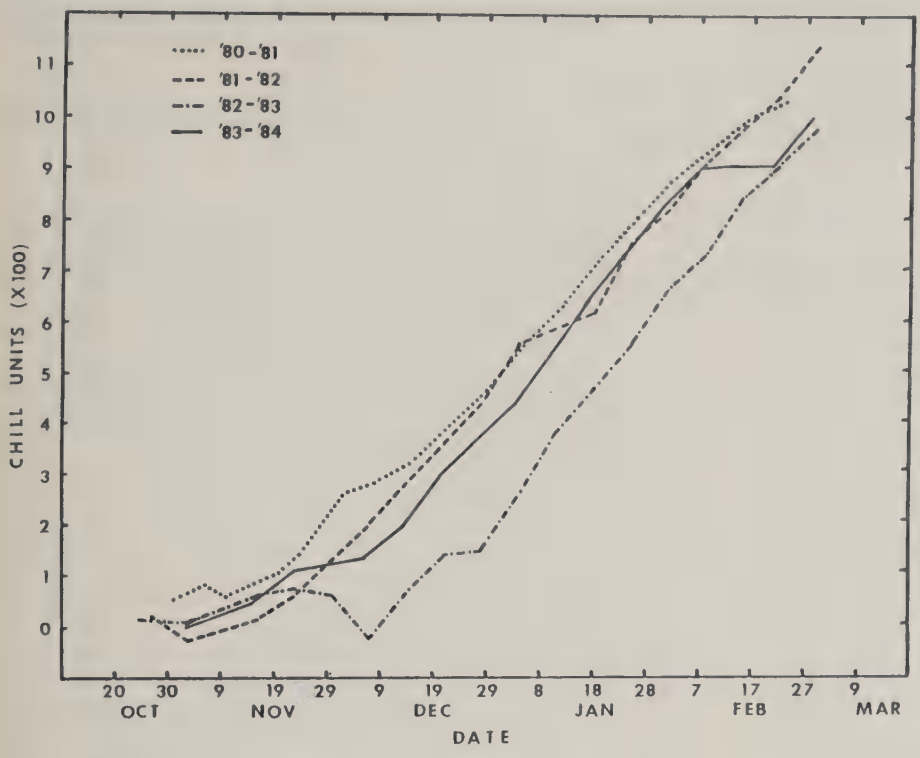

Fig. 1. Chill unit accumulation for 4 pruning seasons.

Table 1. Peach tree mortality as related to pruning date in 'Junegold,' 'Camden,' 'Redglobe,' and 'Redhaven' cultivars, 1980-1984.

Pruning date[z]	No. trees	Cumulative tree mortality 1980-81	1981-82	1982-83	1983-84	% Mortality
October B	57	-[y]	10	10	13	22.8a[x]
November A	58	-	9	15	18	31.0a
B	95	0	8	9	13	13.7b
December A	102	3	10	11	17	16.7b
B	79	2	8	10	13	16.5b
January A	79	2	3	4	7	8.9c
B	71	-	3	4	5	7.0c
February A	73	0	0	1	1	1.4d
B	54	-	0	0	0	0.0d

[z]A = pruned between the first and 15th day of the month; B = pruned after the 15th day of the month.

[y]No trees pruned.

[x]Values followed by the same letter are not significantly different by Chi- Square test, 0.05% level.

Table 2. Peach tree mortality as related to pruning date in four peach cultivars, 1980-1984.

Pruning date[z]		No. trees	Cumulative tree mortality 1980-81	1981-82	1982-83	1983-84	% Mortality
			'Junegold'				
October	B	15	-[y]	4	4	6	40.0
November	A	15	-	3	3	4	26.7
	B	21	0	2	2	3	14.3
December	A	28	0	3	3	5	17.9
	B	21	0	2	2	3	14.3
January	A	21	2	2	2	3	14.3
	B	19	-	0	0	0	0.0
February	A	21	0	0	0	0	0.0
	B	15	-	0	0	0	0.0
			'Camden'				
October	B	14	-	4	4	4	28.6
November	A	14	-	1	3	4	28.6
	B	24	0	5	5	6	25.0
December	A	25	1	3	3	5	20.0
	B	19	1	2	2	2	10.5
January	A	17	0	0	0	0	0.0
	B	17	-	2	2	3	17.7
February	A	16	0	0	0	0	0.0
	B	13	-	0	0	0	0.0
			'Redglobe'				
October	B	15	-	1	1	1	6.7
November	A	15	-	1	3	3	20.0
	B	25	0	0	1	2	8.0
December	A	24	0	0	1	3	12.5
	B	20	1	2	3	5	25.0
January	A	21	0	1	1	3	14.3
	B	19	-	0	1	1	5.3
February	A	18	0	0	1	1	5.6
	B	14	-	0	0	0	0.0
			'Redhaven'				
October	B	13	-	1	1	2	15.4
November	A	14	-	4	6	7	50.0
	B	25	0	1	1	2	8.0
December	A	25	2	4	4	4	16.0
	B	19	0	2	3	3	15.8
January	A	20	0	0	1	1	5.0
	B	16	-	1	1	1	6.3
February	A	18	0	0	0	0	0.0
	B	12	-	0	0	0	0.0

[z]A = pruned between the first and 15th day of the month; B = pruned after the 15th day of the month.

[y]No trees pruned.

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EFFECT OF NEMATODE INDUCED STRESS ON ETHYLENE PRODUCTION IN PEACH TREES

Melissa B. Riley and George E. Carter, Jr.[1]

The presence of either ring (Criconemella xenoplax) or root knot (Meloidogyne sp.) nematodes or both is important to peach growers in South Carolina because these organisms increase the susceptibility of peach trees to cold injury and certain diseases that cause sudden collapse and death of trees. The entire complex of factors that leads to premature death of peach trees has been called peach tree short life. In the 1970's a 10-point program was developed for reducing the incidence of peach tree short life which included post-plant fumigation for nematode control (Brittain and Miller, 1978). With the suspension of 1,2-dibromo-3-chloropropane (DBCP) use in 1979, it became difficult to control the nematodes which had been associated with peach tree short life. The introduction of new effective nematicides has become unlikely, so recent research has focused on obtaining a better understanding of the interaction between nematodes and peach trees and on identifying a rootstock resistant to root-knot and ring nematodes.

An increase in the production of ethylene has been related to various biotic and abiotic stress factors. Glazer, Orion and Apelbaum in Israel have studied the interrelationships of ethylene production, gall development and Meloidogyne infections in tomato plants (Glazer et al., 1983). In susceptible tomato plants ethylene was produced at eight times the rate observed in resistant tomato plants 12 days after inoculation. It has been shown that the immediate precursor of ethylene, 1-aminocyclopropane-1-carboxylic acid (ACC) is transported from waterlogged tomato roots to leaves (Bradford and Yang, 1980). ACC was higher in leaves of tomato plants infected with root-knot nematodes when compared to controls (Glazer et al., 1984). A reversal of nematode-induced growth retardation and gall development was noted when inhibitors of ethylene production or inhibitors of ethylene action were applied to tomato plants (Glazer et al., 1984, Glazer et al., 1985a). Increased ethylene production in response to Meloidogyne infections has been noted for cabbage, pea, carrot, cucumber and carnation plants (Glazer et al., 1985b).

No research has been reported on increased ethylene or ACC production as a response to other nematode genera. This study was initiated to determine if increased ethylene or ACC production in response to root knot or ring nematodes in peach trees could be observed and if it could be related to the resistance of peach rootstocks to nematodes.

MATERIALS AND METHODS

Field Plot. Lovell seedlings were planted April 1983 in 24 lysimeter tanks approximately 5 feet in diameter and 4 feet deep located at the Sandhill Research and Education Center near Columbia, South Carolina. Tanks were fumigated prior to planting with methyl bromide March 1983 at 1.5 lb/tank. In September 1983 the soil in eight tanks was infested with 6000 Criconemella xenoplax and the soil in eight other tanks was infested with 6000 Meloidogyne incognita race 3 nematodes. The check consisted of eight trees. Approximately 100 leaves were taken at random throughout the canopy of available trees June 1986 and placed in a freezer until analyzed for ACC. There were five check, six ring nematode, and four root knot nematode treatment trees remaining at that time.

ACC Extraction Procedure. The extraction of ACC from leaves and other plant material has been previously reported (Chen and Patterson, 1985, Larsen et al., 1986). Leaf material was homogenized with 80% ethanol (2 ml/g fresh weight) then centrifuged. A Brinkman polytron was used to homogenize samples. The supernatant was assayed according to the procedure of Lizada and Yang (1979). ACC was converted to ethylene by a reaction of leaf extract (0.6 ml) with 5 mM mercuric chloride (0.2 ml) and oxidizing reagent (0.2 ml). The oxidizing reagent consisted of 5% sodium hypochlorite (Chlorox) and 10 N sodium hydroxide (2:1, v/v) which was combined immediately prior to use. A gas sample of one ml was removed from the sealed reaction vial one hour after the addition of the oxidizing reagent and injected into the gas chromatograph.

Gas Chromatography. The concentration of ethylene in gas samples was determined using a Varian 3400 gas chromatograph connected to a Vista 600 chromatography data system. The gas chromatograph was equipped with a flame ionization detector and a 2 m X 2 mm (inner diameter) column packed with activated alumina 60/80 mesh. The column, injector and detector temperatures were 100, 150, and 250 C, respectively. The nitrogen carrier gas flow rate was maintained at 30 ml/minute. The air and hydrogen flow rates were 300 and 30 ml/minute, respectively. The retention time for ethylene was 1.4 to 1.5 minutes.

RESULTS AND DISCUSSION

When 80% ethanol was used as an extracting solvent a low level conversion of ethanol to ethylene was observed. Various other extracting solvents including 50% ethanol, 50% isopropanol,

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50% acetone, 5% sulfosalicylic acid, 50% methanol and 80% methanol were tested; and 80% methanol was found to be a suitable replacement for 80% ethanol. More consistant results were also obtained when samples were placed on ice following the addition of the oxidizing reagent.

Low recovery of initial ACC spikes from leaf extracts indicated that incomplete oxidation of ACC to ethylene was occurring because leaf extracts contained many compounds which were being oxidized in addition to ACC. When the final mercuric chloride concentration in the reaction mixture was increased from the 1 to 5 mM and the amount of oxidizing reagent was increased from 0.2 to 0.3 ml a recovery of 82% of the ACC spike was obtained.

The results of the ACC analysis in leaves of nematode infected trees are shown in Table 1. When general linear model LSD's were run on the data a significant difference was noted between root-knot infected trees and both the ring infected and the check treatments. The ring infected and check treatments were not significantly different. Further research is being conducted to determine if differences in ACC production exist between different treatments and if ring nematodes actually have no effect on ACC production as is indicated by these preliminary results.

Table 1. ACC content related to nematode infection in peach trees.

TREATMENT	SAMPLE TREE	nM/G FRESH WEIGHT
Check	# 2	0.19
	5	0.27
	14	0.17
	16	0.21
	22	0.19
		AVG 0.21 nM/g
Ring	4	0.14
	10	0.19
	11	0.29
	13	0.16
	18	0.17
	23	0.19
		AVG 0.19 nM/g
Root Knot	12	0.41
	17	0.22
	20	0.24
	24	0.37
		AVG 0.31 nM/g

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DIFFERENCES IN LEUCOSTOMA TOLERANCE AND COLD HARDINESS AMONG DIVERSE PEACH GENOTYPES

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INTRODUCTION

Cytospora canker, caused by Leucostoma cincta, is the most serious disease reducing peach tree life in Michigan. Symptoms include cankering of the trunk and branches, branch dieback, and progressive weakening and possibly death of the tree. Leucostoma is a wound parasite which enters through dead and dying tissues. Pruning cuts which do not callus properly are ideal Leucostoma entry sites. Winter injury which results in dead and dying tissue predisposes the tree to Leucostoma invasion (Dhanvantari, 1978). Leucostoma-infected trees are commonly observed to be more susceptible to winter injury than healthy trees. Usually the combined influence of low temperature stress and canker is greater than the effect of either acting along.

There has been no large-scale effort to identify genetic resistance or tolerance to Leucostoma in a broad-based population of peach (Layne, 1984). Cultivars do, however, differ in their level of tolerance to Leucostoma but no highly tolerant selections have been identified (Dhanvantari and Dirks, 1983; Luepschen et al., 1975). This is not surprising because the existing level of Leucostoma resistance in North American germplasm is quite low. Most of the peach cultivars are derived from the repeated mating of 'J. H. Hale' and 'South Haven' material. 'J. H. Hale' and its maternal parent 'Elberta' have been rated among the most susceptible cultivars (Dhanvantari and Dirk, 1983; Luepschen et al., 1975). Various inoculation techniques have been investigated to screen for Leucostoma resistance, but no major screening programs have been done (Luepschen, 1981; Scorza and Pusey, 1984).

Because cold injury predisposes the tree to Leucostoma infection, it is necessary to know the hardiness of the inoculated tree. Indications are that well-acclimated, hardy cultivars not only have reduced infection but may have an increased capacity to combat disease progression. The objectives of this study were to evaluate a broad collection of peach germplasm for its winter hardiness in the Michigan climate and evaluate Leucostoma disease development to identify useful genetic material to breed for Leucostoma tolerance.

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MATERIALS AND METHODS

In the spring, 1984, open-pollinated peach seedlings from 19 clones of diverse background (Table 1) were planted at the Horticultural Research Center, East Lansing, Michigan. These seedlings represent half-sib families with progeny numbers ranging from 2 to 73. On October 22, 1985, a 2-year-old branch on each seedling was inoculated with 20 ml of 10^7 Leucostoma spores/ml derived from isolates collected at Clarksville and Hartford, Michigan. A wound-freezing inoculation technique developed by Scorza and Pusey (1984) was followed. A second branch on each seedling was inoculated the following day.

On November 21, two 2-year-old shoots were collected from each seedling and from 13 1-year-old 'Redhaven' trees, placed in a hardiness chamber, and chilled to a critical temperature of -22 C. One week later, the cambium from each shoot was examined and rated as dead or alive. Three additional hardiness evaluations were made on January 9, February 24, and April 4, 1986.

On May 19, when the trees were beginning to leaf out, all the inoculated branches were measured for canker length (length of necrotic area distal to the point of inoculation), branch diameter, and rated for branch health (0 = dead, 1 = severe wilting, 2 = weak growth and slight wilting, 3 = healthy).

Table 1. Parents of the 19 clones used in the Leucostoma/cold hardiness experiment.

Clone	Parents
Babygold 8	PI35201 x Ambergen
Canadian Harmony	Redskin x Sunhaven
Elberta	Chinese Cling (open-pollinated)
Glohaven	(J. H. Hale open-pollinated x Kalhaven
Harken	Redskin x Sunhaven
Loring	Frank x Halehaven
Newhaven	Redhaven x Fairhaven
Reliance	(Minn. (PH04559 x Meredith) open-pollinated
Red Hale	Unknown
Yennoh	Plant Introduction from Russia
B8-11-147	(K82 x Sunrise) x [Red C x NJ191) x Okinawa]
B8-20-171	(5110417 x Ta Tao 3) x C2R31T45
B8-21-20	Orange Cling x RR65-1
C2-28-89	Kasna Dupnishka open-pollinated
C4-11-97	peach x almond
NY257	Honeydew Hale x Jefferson
NJN69	(NJN55 x NJC68) x Marzochella
NJ672017002	(PI35321 x Cherryred) x Prunus kansuensis
RR37-15	NJ174 x Prunus kansuensis

RESULTS AND DISCUSSION

Differences between half-sib families and among progeny within half-sib families were significant for all traits measured (Table 2). Canker length was positively correlated with canker length/cross-sectional area (Table 3). Both canker length and canker length/cross-sectional area were negatively correlated with canker rating where: 0 = dead, 1 and 2 = wilting, and 3 = healthy. Canker length and canker length/cross-sectional area were also negatively correlated with cold hardiness on all the four sampling dates, confirming the association between increased canker development and reduced hardiness. The sum of the hardiness values from the four dates was positively correlated with hardiness values on each of the individual dates.

Progeny mean values for canker length ranged from 4.81 cm to 18.19 cm (Table 4). Progeny from C2-28-89 which had the longest canker length also had the greatest canker length/cross-sectional area plus more dead branches than any other half-sib family. Progeny from 'Reliance' had the smallest canker length and a low canker length/cross-sectional area; however, many of the branches were wilting and would eventually die. In contrast, progeny from 'Yennoh' which had a small mean canker length and the smallest canker length/cross-sectional area, had healthy branches which showed no signs of wilting.

Because the half-sib families had unequal progeny numbers, mean comparisons were not possible. However, when the half-sib families were ranked for mean canker length, the rankings agreed with previous rankings in the literature (Table 5). In general, 'Reliance' and 'Harken' are most tolerant to Leucostoma infection, 'Canadian Harmony' is intermediate, and 'Elberta' and 'Loring' are the most susceptible. The similarity in ranking between the open-pollinated progeny means plus the cultivar data suggest that the differences in Leucostoma tolerance observed in this study are heritable.

Although 'Reliance' is considered to have less canker growth than other cultivars, it does get canker under field conditions. A canker rating of 1.58 indicating wilting and eventual death of the inoculated branches confirms the destructive effect of Leucostoma on 'Reliance.' Although there were only three progeny of 'Yennoh,' the six branches which were inoculated showed no symptoms of wilting. It is possible that 'Yennoh,' a plant introduction from Russia, has a higher tolerance to Leucostoma infection than has been previously found in the U. S. germplasm.

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Table 2. Mean squares and F values for Leucostoma infection and cold hardiness in a population of 693 open-pollinated peach seedlings from 19 cultivars.

Source	df	Canker length		Canker length/ x sectional area		Canker rating		Cold hardiness	
		M.S.	F	M.S.	F	M.S.	F	M.S.	F
Half-sib family	18	535.80	5.66**	176.30	4.34**	14.38	8.20**	17.84	15.31**
Progeny within half-sib family	647	94.57	3.45**	45.23	3.56**	1.75	2.37**	1.17	2.44**
Error	652	27.41		12.70		0.74		0.47	

Table 3. Correlation coefficients between Leucostoma infection and cold hardiness in a population of 693 open-pollinated peach seedlings from 19 cultivars. Cold hardiness was evaluated on the following dates: X_1 = 11/20/85, X_2 = 1/9/86, X_3 = 2/24/86, X_4 = 4/3/86[y].

Trait	Canker[z] rating	Canker length	Canker length/X sect. area	X_1	X_2	X_3	X_4	X_5
Canker rating	1.00							
Canker length	-0.42**	1.00						
Canker length/ X sect. area	-0.34**	0.60**	1.00					
X_1	0.05	-0.14**	-0.06	1.00				
X_2	0.15**	-0.16**	-0.06	0.11**	1.00			
X_3	0.19**	-0.16**	-0.06	0.09*	0.34**	1.00		
X_4	0.18**	-0.17**	-0.19**	0.04	0.16**	0.14**	1.00	
$X_5 = X_1+X_2+X_3+X_4$	0.26**	-0.26**	-0.15**	0.35**	0.70**	0.70**	0.61**	1.00

[y]The critical temperatures were: -22 C, -29.5 C, -28 C, -23 C.

[z]Visual rating: 0 = dead, 1 = severe wilting, 2 = weak growth and slight wilting, 3 = healthy

Table 4. Mean values for Leucostoma infection and cold hardiness from open-pollinated progeny of 19 peach clones.

Clone	No. of progeny	Canker length (cm)	Canker length/ X sect. area	Canker rating[x]	Cold hardiness[y]
Reliance	6	4.81	1.60	1.58	2.92
Yennoh	3	5.30	1.46	3.00	3.50
Newhaven	2	6.20	1.65	2.50	2.75
B8-11-147	30	7.71	3.11	2.10	3.16
NJ672017002	26	8.50	4.03	2.19	3.77
Babygold 8	38	8.81	2.92	1.96	3.54
Glohaven	3	9.03	4.08	2.83	2.83
Harken	30	9.92	3.46	1.30	3.03
B8-20-171	80	9.96	3.91	0.92	3.24
NJ257	63	10.27	3.45	0.88	2.92
Canadian Harmony	58	10.93	3.94	0.86	2.56
Elberta	22	10.95	4.55	0.88	3.38
B8-21-20	53	11.03	4.38	1.15	3.00
C4-11-97	26	11.32	8.38	1.13	3.10
Red Hale	32	11.41	3.03	1.03	3.45
Loring	56	13.36	5.78	0.85	2.46
NJN69	63	14.87	5.35	0.89	2.33
C2-28-89	60	18.19	5.97	0.53	1.65

[x]Visual rating: 0 = dead, 1 = severe wilting, 2 = weak growth and slight wilting, 3 = healthy.

[y]Each value represents the sum of four runs (0 = dead cambium, 1 = healthy cambium).

Table 5. A comparison of published Leucostoma rankings from wound-freeze inoculated peach cultivars compared to rankings from open-pollinated progeny.

Cultivar	Open-pollinated progeny[v]	Scorza and Pusey, 1984[x]	Luepschen et al. 1975[x]	Dhanvantari and Dirks 1983[y]
Reliance	4.81	2.2 a[z]	-	-
Glohaven	9.03	-	-	7.85 a
Harken	9.92	3.8 ab	9.8 ab	6.35 a
Canadian Harmony	10.93	5.5 bc	-	-
Elberta	10.95	10.2 d	12.6 bc	10.7 b
Loring	13.36	7.3 c	14.3 c	-

[v]Length (cm) of necrotic area distal to the point of inoculation on branches of 3-year-old trees.

[w]Length (cm) of necrosis of inoculated wounds minus that of control wounds on young budded trees.

[x]Length (cm) of canker on limbs of ■ 5- to 7-year-old trees; values represent 3-year means.

[y]Length (cm) of necrosis on 1-year-old twigs of 3- to 5-year-old trees; values represent 3-year means.

[z]Means within a column sharing a letter in common are not significantly different, P = 0.05.

SCREENING OF *PRUNUS* GERMPLASM FOR RESISTANCE TO *CRICONEMELLA XENOPLAX* AND PEACH TREE SHORT LIFE

David W. Cain[1], Eldon I. Zehr[2], William R. Okie[3], and Andrew P. Nyczepir[3]

INTRODUCTION

Peach tree short life (PTSL) is a major problem in the southeastern United States. The ring nematode, *Criconemella xenoplax* (Raski) Luc and Raski has been implicated as a major causative factor in inducing PTSL (Nyczepir et al., 1983). Rootstock cultivars 'Lovell' and 'Halford' survive better than others tested on PTSL sites (Dozier et al., 1984). However, both will support high numbers of ring nematodes and may die of PTSL unless nematodes are controlled (Lownsbery et al., 1977). The purpose of this study is to locate a source of resistance to ring nematodes and PTSL in *Prunus* germplasm.

MATERIALS AND METHODS

In 1981, open-pollinated seeds were collected from 143 *Prunus* clones. Lines were chosen to represent as much genetic diversity as possible. Lines included plant introductions from throughout the world, naturalized peaches growing in the southeastern United States, old trees surviving on PTSL sites, commercial rootstocks and related species. All seeds were planted in a fumigated nursery at Clemson, SC, and grown for one year. The dormant seedlings were transplanted to PTSL sites at Elgin, SC, and at Byron, GA. Both sites were chosen because previous orchards had been severely affected by PTSL.

The South Carolina plot consisted of six replicates of eight trees each of 143 *Prunus* lines arranged in a randomized complete block design. The Byron plots consisted of eight replicates containing six trees of 118 *Prunus* lines arranged in a randomized complete block design. A total of 108 lines were common to both locations. Ring nematode counts and tree deaths were recorded in November each year. Soil cores from the base of each tree within a line were collected and thoroughly mixed. Soil samples were stored at 2-5 C until extracted. A 500-cm^3 sample was elutriated using a semiautomatic elutriator (Byrd et al., 1976) and processed using the Jenkins (1964) method. One-fifth of the elutriate was collected and nematode counts were expressed as nematodes per 100 cm^3 of soil. Tree death and cause of death were recorded yearly for each tree.

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RESULTS

Overall, Byron plots had much higher ring nematode counts than Clemson plots (Table 1). Large differences in nematode numbers between cultivars existed at both sites. All cultivars had nematode counts above the 50 nematodes/100 cm^3 soil threshold considered in South Carolina to be economically damaging. Nematode counts exhibited a high degree of variability between replicates, locations and years. Correlation coefficients for Byron nematode counts are: 1983 and 1984, R = .07 (NS); 1984 and 1985, R = .16 (significant at $P = .05$); and 1983 and 1985, R = .06 (NS). Only Tennessee Natural R27 appeared among the lowest counts in both locations while Satsuma Plum had high counts at both locations.

Table 1. *Prunus* seedling lines in field experiments having less than 650 or more than 1300 ring nematodes/100 cm^3 soil at Byron, GA, and less than 100 or more than 500 ring nematodes/100 cm^3 in Clemson, SC.

Line	*C. xenoplax*/100 cm^3 soil	Line	*C. xenoplax*/100 cm^3 soil
Byron			
14DR52	462	SCR5 7	2246
16-167	485	Blue Goose Plum	1733
Redglobe/Lovell	523	Yunnan OP	1678
Halford	527	Angel	1661
Harrow Blood	558	Sary Oilor	1516
Halford Cuttings	610	20-4	1468
Transvaal Yellow	615	Florida 9-4	1437
Tennessee		Satsuma Plum	1415
Natural R27	623	J68-254	1342
SCRS 3	631	49-7	1337
Eagle Beak	635	SCRS 10	1320
Clemson			
De Coosa R27	62	China Flat	974
Opata	67	Satsuma	715
Stanwick	75	Bienvenida	688
Damas 1869	78	Saharan Pur #2	668
Lovell	83	J 67-14	578
Redglobe Cuttings	85	Saharan Pur #1	575
Gaschina Novembre	91	Nemaguard	551
Nooiens Herholdts	94	40-3	542
Tennessee		Soleil D'October	517
Natural R27	98	Killiekrankie	515
GF 655	100	PI Tao	503

Correlations between percent survival and nematode counts at Byron were also very low. The correlations are: survival and 1983 counts R = -.14 (NS); survival and 1984 counts R = -.28 (significant at $P = .05$); and survival and 1985 counts R = .075 (NS).

Tree death due to PTSL was greater at Byron than in South Carolina. Among the 108 lines common to both locations, Byron had a mean tree loss due to PTSL of 29.9% while South Carolina lost 10.1%. There was a good correlation (R = .61) between

deaths among lines at the two locations. For brevity the percent of tree death is not given for all 108 lines. Table 2 gives the overall percent of dead trees averaged over both locations for lines having less than 2.5% or more than 50% tree death. Three lines had no tree losses at either location and 15 lines had less than 2.5% tree loss. Lovell, which is known from previous research to have superior survival on PTSL sites, was among the superior lines.

Table 2. Prunus seedling lines or rooted cuttings having more than 50% or less than 2.5% trees dead from PTSL when averaged over South Carolina and Georgia sites.

Line		% Dead
69	J68-59 P. davidiana FL15-122	86.0
88	3-10 S37 OPx [(RRL x Yunnan)F5]	76.6
18	PI 106062 Killiekrankie	73.7
147	Marianna 2624 cuttings	71.7
95	20-8 S37 OP x [(RRL x Yunnan)F5]	69.4
112	P. davidiana Byron	65.5
87	3-6 S37 OP x [(RR1 x Yunnan)F5]	61.6
4	PI 55776 Yunnan OP	58.8
126	Ark 7768 Boone Co. x Siberian C	55.5
7	PI 65821 Shau Thai Tao Sdlg	54.6
89	3-12 S37 OP x [(RRL x Yunnan)F5]	54.2
54	Siberian C	51.2
81	14DR53 Redleaf Beltsville Md	2.5
37	PI 134151 Travsvaal Yellow	2.4
107	Satsuma Plum P. salicina	2.4
132	SCRS-1 Madison NC	2.3
35	PI 133987 Nooiens Herholdt's	2.1
57	Tennessee Natural R27	2.0
137	Lovell Cuttings	2.0
63	J37-34 Tenn Nat Boyd Nur. TN	1.3
133	SCRS-3 Madison Co. NC	1.2
86	152AI-2RH-2 S37 Sel. NC	1.2
109	Edible Sloe ? spps AL	1.0
70	J68-69 Indian Blood-Plum Peach GA	1.0
55	Tennessee Natural R2	0.0
73	J68-271 Very Old Tree GA	0.0
104	520-9 [(Nemaguard OP)OP]OP	0.0

Among the poorest surviving lines were two lines of Prunus davidiana, several lower chilling lines and several root-knot nematode-resistant lines. Among the better performing lines were several lines originating in the southeastern United States.

Figure 1 compares several of these subgroups to the overall population of lines. Lines having at least one root-knot nematode-resistant parent had poorer survival than the overall population. However, when 13 lines having S37 OP x [(Rutgers Red Leaf x Yunnan) F5] were eliminated, the remaining root-knot nematode-resistant lines had only a slightly higher death than the overall population. Siberian C and lines having Siberian C in their ancestry performed very poorly with almost 85% tree loss in the Byron plots and 29% loss in South Carolina. Several lines originated from a collection made to locate healthy old trees growing on PTSL sites throughout the Southeast. As a group these lines had a much lower tree loss than the overall population.

Tennessee Naturals are derived from peaches originally introduced into South America by the Spanish and have been growing in a semicultivated state in the southeastern United States since the early 16th century. As a group these lines had 7.4% tree loss in Byron and 1.6% in South Carolina. Three lines had no tree loss at either location (Table 2).

DISCUSSION

Field screening for ring nematode resistance is not very promising due to the extreme amount of random variation in nematode counts which cannot be controlled through experimental design, although genetic lines having superior survival can be identified by field screenings. Although lines were not chosen to allow a detailed genetic analysis, large differences in tree losses occurred among genetically related groups, indicating a degree of genetic control. The superior performance of Tennessee Naturals and other lines originating in the Southeast indicates that some natural selection for tolerance to PTSL conditions may have occurred in these populations. While root-knot nematode-resistant lines as a group were above average in susceptibility to PTSL, at least one root-knot-resistant line, 520-9, a Nemaguard O.P. F3 seedling, has had no tree losses to date. A second root-knot-resistant line, 152 AI-2RH-2 derived from S37, has only 1.2% dead trees. This experiment will continue to monitor tree losses to determine if any genetic lines will continue to exhibit good survival on PTSL sites.

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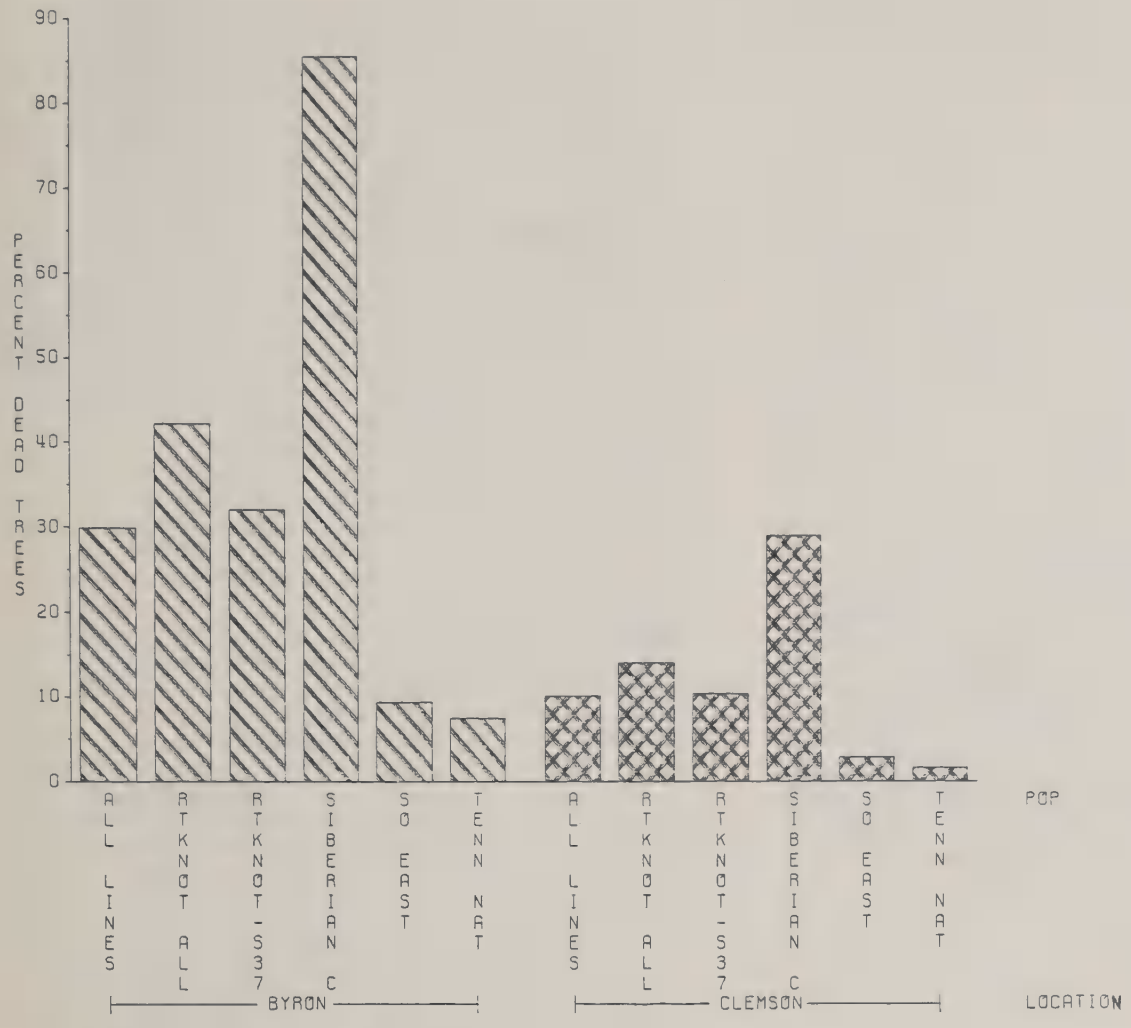

Figure 1. Percent Trees Dead of PTSL at Byron, GA. and Elgin, S.C.

POPULATION DYNAMICS OF RING NEMATODES AND PEACH TREE SHORT LIFE IN NORTH CAROLINA

David F. Ritchie[1]

INTRODUCTION

Peach tree short life (PTSL) is a disease complex characterized by the failure of trees, which were apparently healthy the previous fall, to initiate growth in spring or growth is initiated and the trees or portions of trees suddenly collapse, usually during bloom or leaf development, with subsequent premature death (Ritchie and Clayton, 1981). Numerous interacting factors, both direct and predisposing, have been implicated in the PTSL complex (Carter, 1976; Nyczepir et al. 1983; Reilly et al., 1986; Ritchie and Clayton, 1981; Weaver et al., 1974; Zehr et al., 1976). The ring nematode, Criconemella xenoplax, has been implicated as a predisposing factor causing increased sensitivity to freeze injury (Carter, 1976; Nyczepir et al., 1983; Reilly et al., 1986); bacterial canker, Pseudomonas syringae pv. syringae (Lownsbery et al., 1977; Lownsbery et al., 1973; Weaver et al., 1974); and Cytospora canker, Cytospora cincta and C. leucostoma (English et al., 1982). All three of these, either alone or in combination, are considered to be direct factors leading to tree death in the PTSL complex.

Although investigators have observed an association of C. xenoplax with premature tree death (an example is shown in Fig. 1) and Nyczepir et al. (1983) have demonstrated PTSL can be initiated by adding C. xenoplax to peach trees near planting time, almost no reports exist in which the population dynamics of ring nematode and its association with peach tree conditions have been followed over a period of years. Such an experiment has a great deal of difficulty not only from the length of time over which it should be done but it involves extensive nematode sampling and environmental and tree condition monitoring. Additionally, estimates of nematode populations can vary greatly and methods for determining tree condition can be more subjective than objective.

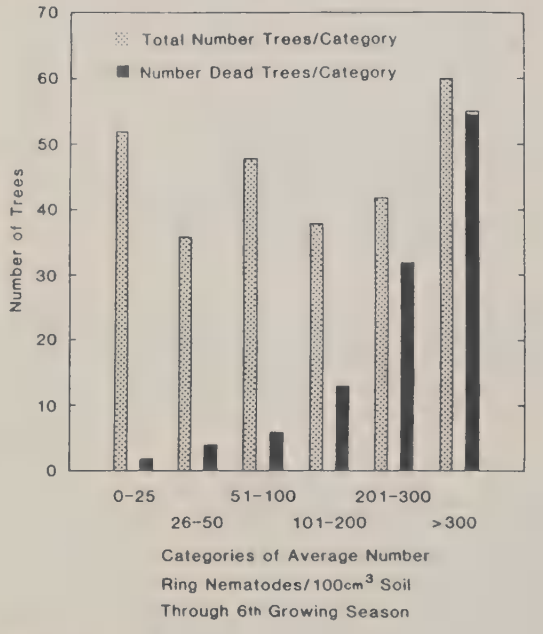

Figure 1. Association between the magnitude of average ring nematode populations during a 4 year period and tree death.

The data to be discussed in this presentation were obtained from three plots located at the Sandhills Research Station, Jackson Springs, NC. The initial objective of these three plots was to evaluate pre- and postplant nematicide treatments. This has allowed for varying ring nematode population levels and tree conditions. However, this approach could also introduce the possibility that the nematicides have an effect not limited solely to the nematode but could by themselves affect tree physiology. Plots F1A and E2B were Lovell budded to Lovell rootstock and Winblo budded to Lovell rootstock, respectively. The trees were planted in March 1980 in typical Sandhills's soil [light, sandy soil (Candor sand) with less than 2% organic matter and a pH of 5.6 to 6.0 during the experiment] which had been broadcast fumigated the previous October with dichloropropene-dichloropropane (D-D). Postplant treatments with fenamiphos (Nemacur 3) were initiated in September 1981 in F1A (end of second growing season) and in September 1982 in E2B (end of third growing season). The third plot, D4W, consisted of Emery budded to Lovell rootstock. Trees were planted in March 1983 in soil similar to that for the other two plots except one-third was treated with 1,3-dichloropropene (Telone II), one-third with fenamiphos (Nemacur 3) at planting, and one-third was not preplant treated. Postplant treatments with fenamiphos were initiated from September 1983 through April 1985.

The overall objective of the following discussion is to present data obtained from these three plots and, primarily through the use of regression models, attempt to better understand the association between ring nematodes and peach tree conditions in the PTSL complex. Parameters compared will be ring nematode populations; cambial browning rating (CBR), which is a subjective rating used to estimate the severity of freeze injury to cambium and bark in the trunk of the tree, on a scale of 0-9 with 0 indicating no browning and 9 indicating death; and the percentage of tree death.

Ring Nematode Populations First Two Years After Planting Trees. The greatest increase in ring nematode populations occurred during the first two years after the trees were planted (Fig. 2). Magnitude of the populations was affected by the preplant soil treatments. When trees were planted in untreated soil (the previous trees had been removed four years earlier), populations increased to an average of 700 larvae/100 cm^3 soil within an 18-month period. By the start of the third growing season (April 1985), populations,

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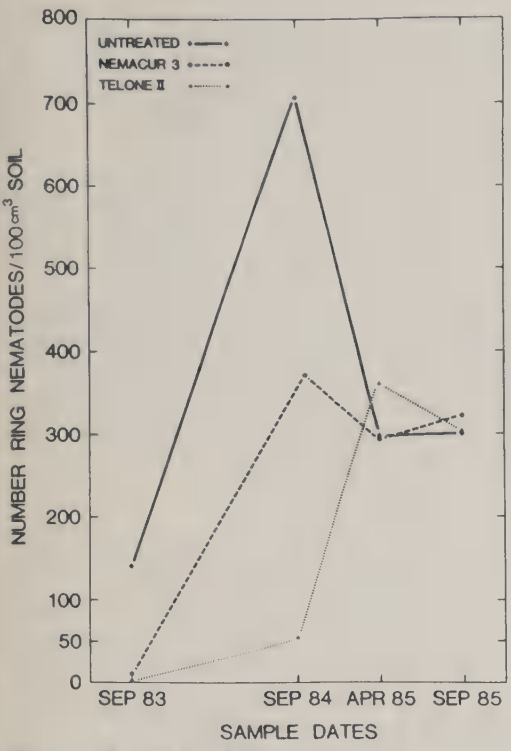
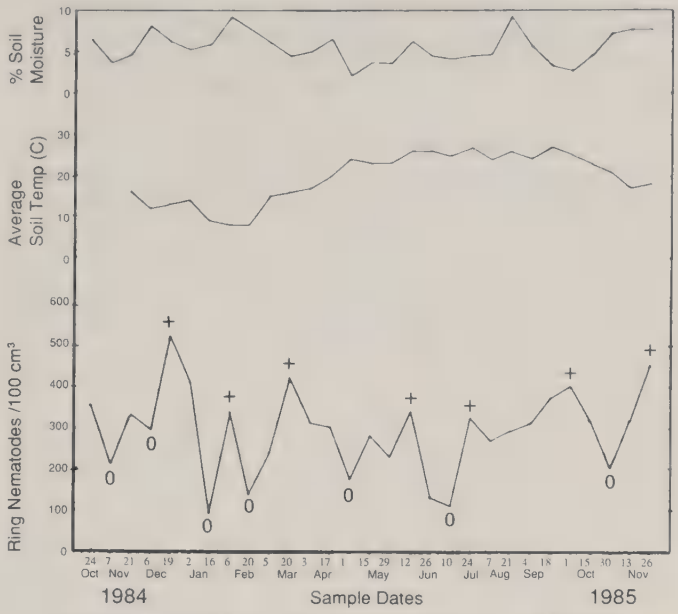


Figure 2. The effect of different preplant treatments on ring nematode populations following planting of peach trees. Trees were planted February 1983.



Peaks designated by "+" are significantly different from preceeding low point designated by "0"; LSD (P = 0.05).

Figure 3. Results of monitoring ring nematode population from Oct 1984 through Nov 1985. Each point is the mean of nine samples taken from the same trees at 2-week intervals.

regardless of the preplant treatment, stabilized at approximately 300 larvae/100 cm^3 soil.

From October 1984 through November 1985 (end of 2nd to end of the 3rd growing season), ring populations and percent soil moisture were monitored every two weeks and soil temperature at 10-12 cm depth was monitored continuously. Nine nematode samples were taken, three from each of the preplant treatment regimes, at each sample period. During this period, there were seven significant (LSD, P = 0.05) peaks in the ring nematode populations. On the average, the ring populations remained relatively constant during this monitoring period and neither soil temperature nor moisture appeared to become limiting (coefficient of determination R^2 < 0.01) (Fig. 3).

Cambial Browning and Ring Nematode Populations. Cambial browning ratings (CBR) were done in late February to mid-March of each year that cambial browning occurred. Cambial browning does not occur every winter but is most likely to occur when temperature extremes are great. Linear regression indicates the severity of cambial browning was positively correlated with an increase in ring nematode populations; however, cambial browning occurred even when ring nematode populations were zero (Fig. 4). This may suggest that cambial browning is indeed an indication of freeze injury and that the degree of severity can be influenced by ring nematode populations. Furthermore, cambial browning does not necessarily mean tree death will occur. The regression of CBR

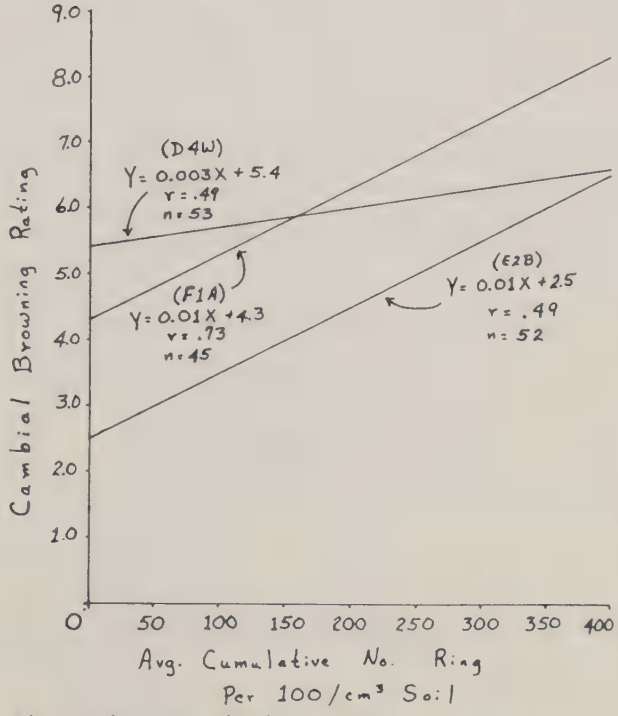


Figure 4. Association between the average cumulative ring nematode population through 1985 and the cambial browning rating in 1985.

on the percent tree death indicates that an average CB rating of 4.1 or greater is required before tree death is likely to start.

Percent Tree Death and Ring Nematode Populations. Regression of % tree death on average cumulative ring nematode populations allows one to answer the question, "What is the population at which tree death is likely to start to occur?" Results shown in Figure 5 indicate that at least for the three plots monitored the injury threshold was between 38 and 83 average cumulative number of ring detected/100 cm^3 soil.

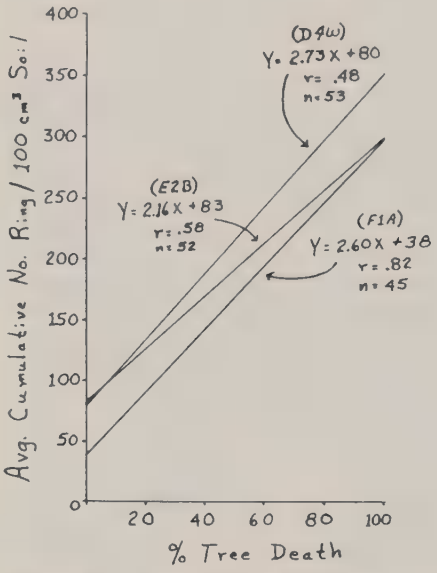


Figure 5. Association between average cumulative ring nematode population through 1985 and % tree death in 1985.

Multiple Regression. Regression models for estimating cambial browning and % tree death are based on average cumulative ring nematode populations and CBR for each of the three plots. The regression models and variables composing the models follow.

Multiple Regression Models

F1A

$$R^2 = .84$$

$$Y_{death85} = -.05X_1 + .07X_2 - .008X_3 + 10.0X_4 - 1.9X_5 - 4.7X_6 + 15.5X_7 - 41$$

$$R^2 = .74$$

$$Y_{CBR85} = -.0004X_1 - .003X_2 - .01X_3 + .02X_4 - 0.1X_5 + 6.2X_6 + 1.7$$

X_1 = ring Sep 81
X_2 = ring Sep 81-Sep 82
X_3 = ring Sep 81-Sep 83
X_4 = ring Sep 81-Sep 84
X_5 = CBR Mar 82
X_6 = CBR Mar 84
X_7 = CBR Mar 85

E2B

$$R^2 = .53$$

$$Y_{death85} = -.02X_1 - .04X_2 - .14X_3 + 4.0X_4 - 2.65X_5 - 17.1$$

$$R^2 = .56$$

$$Y_{CBR85} = -.002X_1 + .004X_2 - .002X_3 + .68X_4 + 1.2$$

X_1 = ring Sep 82
X_2 = ring Sep 82-Sep 83
X_3 = ring Sep 82-Sep 84
X_4 = CBR Mar 84
X_5 = CBR Mar 85

D4W

$$R^2 = .36$$

$$Y_{death85} = -.05X_1 + .25X_2 - .14X_3 + 8.2X_4 - 35.4$$

$$R^2 = .23$$

$$Y_{CBR85} = -.0005X_1 + 4.57X_2 + 5.4$$

X_1 = ring Sep 86-Sep 85
X_2 = ring Sep 83-Apr 85
X_3 = CBR84
X_4 = CBR85

The best "fit" was obtained with the data from plot F1A. Coefficient of determination, R^2, was .84. Using this model, hypothetical situations with ring nematode populations and CBR from 1981 to 1985 were tested for the effects on the % tree death in 1985 (Table 1). The model predicts that the greatest percentage of tree death occurs with an increasing ring nematode population and increasing freeze injury (CBR). An opposite situation resulted in no tree death. If ring was present without freeze injury occurring or freeze injury was moderate and ring was not present, tree death was less than 5%.

Table 1. Hypothetical situation used in the model developed with data generated from F1A.

Situation	S81 (X_1)	S81-S82 (X_2)	S81-S83 (X_3)	S81-S84 (X_4)	CBR82 (X_5)	CBR84 (X_6)	CBR85 (X_7)	% Death (Y)
Increasing ring and freeze injury.....	50	75	150	250	1.0	5.0	7.0	85
Decreasing ring and freeze injury.....	250	150	75	50	7.0	5.0	1.0	0
Ring = 50 with no freeze injury.....	50	50	50	50	0.0	0.0	0.0	0
Ring = 0 with moderate freeze injury.....	0	0	0	0	5.0	5.0	5.0	4
Large ring and no freeze injury.....	200	200	200	200	0.0	0.0	0.0	2
Initial large ring population.....	300	25	10	10	6.0	1.0	1.0	0

Model: $Y = .05X_1 + .07X_2 - .008X_3 + .10X_4 - 1.9X_5 - 4.7X_6 + 15.5X_7 - 41$ $R^2 = .84$.

Reilly et al. (1986) found in a recent study of PTSL and tree physiology, environment, pathogens, and cultural practices that PTSL was related more to high C. xenoplax populations and low soil pH than to pruning. Also, in their study, the primary injury to PTSL trees appeared to be from cold damage. The model for plot F1A presented in this paper would support their hypothesis concerning high C. xenoplax populations and freeze injury.

Using similar models, it may be possible to better estimate which orchards have the potential to incur losses from PTSL. Although cambial browning is an indicator of the injurious effects of temperature, a model with temperatures directly incorporated would probably be more useful.

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IDENTIFICATION AND DISTRIBUTION OF ROOT ROT PROBLEMS IN SOUR CHERRY IN MICHIGAN

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INTRODUCTION

Tree mortality is a problem in numerous commercial cherry orchards in Michigan. Major portions of the root systems are found to be dead when trees are pulled from their planting sites. Symptoms typical of Armillaria root rot are commonly observed on declining trees planted on sandy soil, while symptoms resembling Phytophthora root and crown rot are observed on declining trees on heavy soils.

Our objectives were to determine the incidence and distribution of Armillaria root rot and Phytophthora root and crown on sour cherries in Michigan and to identify the species of Armillaria and of Phytophthora involved.

MATERIALS AND METHODS

During September and October of 1985 and 1986, basidiocarps (mushrooms) of Armillaria were collected from sour cherry, sweet cherry, apple, and peach trees in the western Michigan fruit belt. Basidiocarps were also collected from oak stumps at two locations. To obtain single basidiospore isolates, basidiospores were discharged onto the surface of 1% malt agar. After 12-24 hr, individual germinated basidiospores were transferred to petri plates containing the SR medium of Shaw and Roth (1976). Some 8-12 single spore isolates were obtained from each basidiocarp. Single basidiospore isolates are haploid.

Where basidiocarps failed to release basidiospores, isolations were made directly from basidiocarps. If basidiocarps were lacking, isolations were made from infected woody tissues. Haploid isolates were produced from diploid isolates by inducing somatic segregation on SR medium amended with benomyl (Anderson and Yacoob 1984).

Sexual compatibility among the isolates was tested by pairing haploid isolates on SR medium (Anderson and Ullrich 1979; Ullrich and Anderson 1978). Haploid isolates from each of 33 basidiocarps were paired with themselves to establish tester strains for each basidiocarp. These tester strains were crossed with tester strains from the other basidiocarps and intersterile groups were established. To determine the species of Armillaria in Michigan orchards, tester isolates representative of the intersterile groups in our pool of Michigan isolates were crossed with tester strains provided by J. B. Anderson, University of Toronto, for North American Biological Species I, II, III, V, VI, VII, IX, and X of Armillaria (Anderson and Ullrich 1979).

Isolations for Phytophthora were made from the roots and crowns of trees showing symptoms of Phytophthora root and crown rot in 18 sour cherry orchards. From each tree, no less than 50 tissue sections were placed on a selective medium for Phytophthora (Harris and Bielenin 1986). Identification of isolates was based on vegetative growth rates on cornmeal agar and on sporangia morphology on clarified V-8 juice agar (CV8A) flooded with 1.5% sterile soil extract. Oogonia, antheridia, and oospores were produced on CV8A.

RESULTS AND DISCUSSION

Armillaria was confirmed by isolation from 108 sour cherry, 6 sweet cherry, 1 apple, and 4 peach trees (Table 1). These 37 orchards were planted on sandy soils and were located from New Era to areas north of Traverse City, except for one orchard in Van Buren county, in southern Michigan.

Four intersterile groups were identified among the Armillaria isolates from fruit trees. These four intersterile groups were compatible with tester isolates of North American groups I, III, VI, and VII, respectively. All isolates from oak were compatible with tester isolates of group VI. Isolates in group I correspond to A. ostoyae, isolates in group VI to A. mellea sensu stricto, and isolates in group VII to A. bulbosa (Anderson et al. 1980; Morrison et al. 1985; Wargo and Shaw 1985). Isolates in group III have not been associated with a known taxonomic species. From the 119 orchard trees sampled, A. ostoyae, A. mellea, A. bulbosa, and group III were collected from 89, 26, 1, and 3 trees, respectively. This is the first time A. ostoyae has been identified as a pathogen of fruit trees.

The analysis of the biological species of Armillaria on sour cherry indicates that the species of Armillaria found in Leelanau, Grand Traverse, and Benzie counties is A. ostoyae. In Manistee and Mason counties, group III and A. ostoyae were recovered from orchard trees. In Oceana county, A. mellea was the predominate species of Armillaria present, but A. ostoyae and A. bulbosa were also collected. The single isolate from Van Buren county was A. mellea. Except for one site, only one species of Armillaria was found per orchard.

Phytophthora was isolated from 41 of 58 symptomatic sour cherry trees from 18 orchards in 9 counties of western Michigan. Phytophthora megasperma, P. cryptogea, P. cactorum, and P. syringae were recovered from 17, 4, 2, and 2 orchards, respectively. Often two or three species were recovered from a single orchard. All species were pathogenic to Mahaleb seedlings planted in artificially infested potting media.

Of the species of Phytophthora isolated in this study, P. megasperma and P. cryptogea have been demonstrated to be pathogens of cherry in other

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Table 1. Distribution pattern of biological species of Armillaria isolated from orchard sites in Michigan.

County	Farm	Host	Trees (no.)	Species[a]
Leelanau	A	sour cherry	7	I
	B	sour cherry	3	I
	C	sour cherry	3	I
	D	sour cherry	5	I
		sweet cherry	2	I
	E	sour cherry	1	I
	F	sour cherry	5	I
	G	sour cherry	1	I
	H	sour cherry	1	I
	I	sweet cherry	4	I
	J	sour cherry	1	I
	K	sour cherry	1	I
Grand	L	sour cherry	6	I
Traverse	M	sour cherry	5	I
	N	sour cherry	6	I
	O	sour cherry	3	I
	P	sour cherry	2	I
	Q	sour cherry	5	I
	R	sour cherry	5	I
	S	sour cherry	2	I
Benzie	T	sour cherry	5	I
	U	sour cherry	1	I
	V	sour cherry	1	I
Manistee	W	sour cherry	1	III
	X	peach	4	I
		sour cherry	1,1	I,III
	Y	sour cherry	2	I
	Z	oak	5	VI
Mason	AA	sour cherry	1	III
	BB	sour cherry	2	I
		apple	1	I
Oceana	CC	sour cherry	5	VI
		oak	1	VI
	DD	sour cherry	1	VI
	EE	sour cherry	6	VI
	FF	sour cherry	4	I
	GG	sour cherry	4	VI
	HH	sour cherry	2	VI
	II	sour cherry	5	VI
	JJ	sour cherry	2	VI
	KK	sour cherry	1	VII
Van Buren	LL	sour cherry	1	VI

[a]North American biological species of Armillaria based on the classification system of Anderson and Ullrich (1979).

states (Mircetich and Matheron 1976; Wilcox et al. 1985; Wilcox and Mircetich 1985). Our results provide additional data for implicating P. cactorum and P. syringae in crown and root rot of cherry. Previously, P. cactorum and P. syringae were recovered from diseased sweet cherry trees in single orchards in California (Wilcox and Mircetich 1985), and P. syringae was recovered from cherry soils but not from cherry tissues in New York (Wilcox et al. 1985).

CONCLUSION

Armillaria root rot and Phytophthora root and crown rot were confirmed by isolation and identification of the respective pathogens as significant causes of tree mortality in Michigan sour cherry orchards. Both diseases were widespread in occurrence. These data provide justification for continuing research directed toward the eventual control of these diseases.

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ELISA INDEXING SURVEY FOR PRUNUS NECROTIC RINGSPOT VIRUS IN WEST VIRGINIA PEACH ORCHARDS.

J. G. Barrat[1/] and L. A. Schwartz[2/]

INTRODUCTION

Enzyme-linked immunosorbent assay (ELISA) indexing of peaches for Prunus necrotic ringspot virus (PNRSV) during the past 4 years indicated the presence of this virus in variable quantities of infected trees in the orchards tested. PNRSV reduces the growth and yield of trees within the orchard (Barrat & Otto, 1985; Milbrath, 1957; Parker, et al., 1959; Pine, 1984) and poses a danger for commercial growing of stone fruits (Topshiiska, 1982). Transmission of PNRSV by insects, mites or nematodes in this country has not been recorded. However, transmission of PNRSV by pollen has been reported in several stone fruits; rapid spread in sour cherry in Midwestern and Northeastern states and slow spread in sweet cherry, prune and cling peach in the Western states (Nyland, et al., 1976). Little is known about the source of PNRSV infections, its persistence in the orchard and its spread by pollen in the freestone peach orchards of the mid-Atlantic fruit growing region.

Orchard spread of PNRSV varies within the Prunus species (Cameron, et al., 1973). The purpose of this paper is to present information on 4 years of ELISA indexing of peach trees to clarify some factors about source of infection, persistence and spread of PNRSV.

MATERIALS AND METHODS

Groups of peach trees from many orchards and varieties were indexed in 1983, 1984, 1985 and 1986 using the ELISA procedure of Clark and Adams (1977). The PNRSV antiserum was obtained from American Type Culture Collection (ATCC), Rockville, Maryland 20850.

Tissue samples were taken from terminal leaves on vigorously growing shoots located on a major scaffold limb in the central portion of the tree. Samples were collected and processed immediately or refrigerated overnight. Leaf samples were ground in a Brinkmann Homogenizer (Brinkmann Instruments Company, Westbury, New York 11590) in a 1:20 ratio (w/v) with PBS-Tween PVP buffer. Enzyme-globulin conjugates were used at 1:500 v/v. Wells of microtiter plates were coated with 200 ul of globulin at 5 ug/ml. Reaction intensity was measured photometrically at 410 nm with A Dynatech Microelisa Mini Reader MR590 (Dynatech Laboratories, Inc., Alexandria, VA 22314). Controls included 2 healthy peach leaf samples and 2 diseased peach leaf samples per 60 sample wells. ELISA reactions which gave absorbances equal to or greater than twice the average reading for healthy control samples were regarded as positive (Lister, et al., 1980).

The survey compared 5 age groups of trees: #1) first-year planted trees, #2) 1- to 4-year-old trees (which had not bloomed), #3) 5- to 8-year-old trees, #4) 9-year-old and above trees, and #5) 5-year-old and above trees (which had bloomed in all but the 1985 season). At least 1 year passed between comparative indexings.

#1) First-year planted trees (222 trees, 8 orchard sites, 5 varieties: Early Sunhigh, Sunhigh, Redskin, Rio Oso Gem, and a mixture) were initially ELISA indexed less than 2 months after orchard planting.

#2) One- to four-year-old trees (791 trees, 29 orchard sites, 13 varieties: Garnet Beauty, Redhaven, Early Sunhigh, Sunhigh, Glohaven, Early Loring, Loring, Blake, Redskin, Rio Oso Gem, Baby Gold 5, Baby Gold 7, and a mixture) were indexed at least 1 year following initial indexing. Because of juvenile age and the complete freeze-out in 1985, fruit buds did not produce pollen on these trees.

#3) Five- to eight-year-old trees (467 trees, 17 orchard sites, 8 varieties: Earliglo, Redhaven, Sunhigh, Glohaven, Loring, Blake, Cresthaven and Redskin) were tested. Peach trees in this age group were vigorous, flowered normally, produced ample fruit buds and pollen (except in the 1985 season).

#4) Nine-year-old and above trees (83 trees, 3 orchard sites, 2 varieties: Sunhigh and Loring) also flowered normally and produced fruit buds (except in the 1985 season).

#5) The 5-year-old and above trees were composed of 550 trees from groups #3 and #4.

RESULTS AND DISCUSSION

PNRSV indexed trees showed differences in the percent of initial infection in the orchard test blocks. Infection ranged from 9.5% to 22.9% in the 5 age groups of orchard trees. When grouped in 4-year increments, younger trees had less infection than older trees.

First-Year Planted Trees

No. trees tested	1st test no. pos.	% pos.	2nd test no. pos.	% pos.
222	21	9.5	20	9.0

Indexing Change			
- to +	%	+ to -	%
1	0.4	2	0.9

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The first-year planted trees gave an initial 9.5% positive response to PNRSV infection. These trees were retested at least 1 year later and showed 9.0% positive response. The indexing indicated a change of 0.4% from a negative to a positive response and 0.9% from a positive to a negative response.

Since trees in this category had been planted less than 2 months when initially tested, and since no method is known which would permit transmission of PNRSV to occur within such a short period of time, the PNRSV-positive tested trees were considered to have been infected when obtained from the nursery. The second testing, a year later, indicated approximately the same number of infected trees with no evidence of spread of the virus. The change of index readings was a low percentage and within the range of experimental error (Setula et al., 1986).

One- to Four-Year-Old Trees

No. trees tested	1st test no.	1st test pos.	% pos.	2nd test no.	2nd test pos.	% pos.
791		116	14.7	114		14.4

Indexing Change			
- to +	%	+ to -	%
8	1.0	10	1.3

The first testing of this group showed 14.7% of the trees infected with PNRSV. The second testing showed 14.4% with a positive response. This was an indexing change of 1.0% from negative to positive and 1.3% from positive to negative.

This age category had no opportunity for fruit bud formation and pollen formation. No measurable increase in infection occurred. That PNRSV transmission by insects, mites or nematodes was not detected corresponds to data for other stone fruits in a 1- to 4-year-old age group (Cameron et al., 1973; George & Davidson, 1963).

Five- to Eight-Year-Old Trees

No. trees tested	1st test no.	1st test pos.	% pos.	2nd test no.	2nd test pos.	% pos.
467		106	22.7	106		22.7

Indexing Change			
- to +	%	+ to -	%
8	1.7	8	1.7

In this group 22.7% PNRSV infection was found to be present in both tests. The indexing change in each case was 1.7%.

These trees flowered normally and produced ample fruit buds and pollen for possible virus transmission. However, the data indicated a low percentage (1.7%) of indexing change from negative to positive (also within experimental error bounds) showing no significant transmission of PNRSV in peach orchards of this age group.

These findings do not correspond to data for cherry indicating the number of infected trees to increase geometrically for the first 4 years of bloom (Cameron et al., 1973; Gilmer & Way, 1960).

Nine-Years-of-Age and Older Trees

No. trees tested	1st test no.	1st test pos.	% pos.	2nd test no.	2nd test pos.	% pos.
83		19	22.9	21		25.3

Indexing Change			
- to +	%	+ to -	%
3	3.6	1	1.2

This group exhibited 22.9% initial PNRSV infection and 25.3% in the second test. The indexing change was 3.6% from negative to positive and 1.2% from positive to negative. The indexing change remained within experimental error limits.

The initial infection level in this group was similar to the 5- to 8-year-old trees but higher than in younger trees. The higher rate of initial infection was probably because the trees were planted before there was much attention paid to virus infection in nursery stock. In recent years greater attention has been given to virus content in budwood sources and propagation procedures in the nurseries. There was indication that the number of infections came with the nursery stock and remained stable during the years. These trees had ample opportunity to exhibit spread by pollen within the orchards. Schmitt, et al. (1977) reported that infections in California cling peach increased from 4% at planting to 27% in 9 years, 48.5% in 12 years and 82% in 14 years resulting from pollen transmission. The freestone peach varieties in this study did not exhibit this increase in infection. The increase in indexing change remained low and within the range of experimental error.

Five-Years-of-Age and Older trees

No. trees tested	1st test no.	1st test pos.	% pos.	2nd test no.	2nd test pos.	% pos.
550		125	22.7	127		23.1

Indexing Change			
- to +	%	+ to -	%
11	2.0	9	1.6

In the 5-year-old and above group, 22.7% of the trees initially tested positive. The second test showed 23.1% of the trees positive. The indexing changes of 2.0% trees from negative to positive and 1.6% trees from positive to negative were within the range of experimental error.

The data from this group shows the comparison of trees which had not bloomed (1- to 4-year old trees) with trees that had bloomed. The

rates of indexing change were only slightly higher in the older trees.

The major source of PNRSV in freestone peach varieties in this area may be attributed to infected nursery stock. Trees infected with the virus remain infected for their lifetime. PNRSV has not been shown to spread by any means in this survey. Contrary to reports of rapid dissemination of PNRSV by pollen in stone fruits in other areas, PNRSV was not found to spread readily by pollen in peaches in the mid-Atlantic fruit growing area. The indexing change is such a low percentage that transmission may occur at a percentage less than the experimental error or not at all. Since pollen collected from infected freestone peach trees which indexed positive by ELISA for PNRSV (Barrat, unpublished data), the potential was present for pollen transmission in the orchards, although transmission was not indicated in this study.

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SOME FACETS OF THE ECOLOGY OF PRUNUS NECROTIC RINGSPOT VIRUS IN PEACH TREES IN SOUTH CAROLINA.

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Literature on infections by prunus necrotic ringspot virus (PNRSV) of peach trees growing in the southeastern U.S.A. is scarce (Wells et al.,1986). A considerable volume of work has been completed on the ecology of the virus in cherry, and work on other Prunus species including peach has been completed in some western and northern states. However, climatic differences between areas of the U.S. and differences among the growth habits of peach and other Prunus species may make direct extrapolation of the information from one region to the other invalid.

In preliminary work with PNRSV in peach trees in S. Carolina we have examined the localization of the virus within the tree with the object of maximizing the likelihood of detecting the virus by ELISA or other assays and providing information on the development of systemic infection within the tree. We have also collected data on the rate of re-infection of a healthy planting from external sources and have anecdotal information on the potential rate of spread of the virus within a variety once a focus of infection has been established.

Using direct, double antibody sandwich ELISA, with antibodies prepared from antiserum to Fulton's strain G of PNRSV (ATCC PVAS 22, 1982), we have detected the virus in both blossom and leaves. Trees were sampled over a 2 year period. Samples of blossom were taken, and one month later a sample of leaves was taken. This sampling procedure was repeated in the second growing season. In trees where the virus was detected in the blossom, the leaf sample was also usually found to contain the virus. Exceptions to this generalization exist. In a few trees infections detected in the blossom were not detected in leaves developing in the same year, but the virus was usually detected in the blossom and leaves in the second growing season. However, in eight trees in which the virus was detected in blossoms in the first year, it was not detected in any subsequent assay.

Certain infected trees identified during this work were subjected to a detailed examination to determine the distribution of the virus within the tree. Plans of the individual trees were drawn, leaf samples taken, and the sample sites recorded on the plan. Trees were identified in which the virus was restricted to a single scaffold limb, to individual branches on a scaffold limb or to individual leaves on a single budstick. Examination of these same individual trees during the second year of growth revealed that with some trees the infection had become systemic, whereas with others the infection was still restricted to specific areas of the tree. Despite this localization within the tree, we have found that by assaying a combined sample composed of samples of either leaves or flowers from each quadrant of the tree we have been able to detect the virus with a high degree of reliability.

A planting of virus-free peach trees (170 trees in an area of 1.7 acres) was established in 1978 as part of the South Carolina Peach Tree Certification Scheme. The trees in this planting have since been assayed twice a year for the presence of PNRSV in order that infected trees can be eliminated and the budwood and seed supplied from this block be maintained free of PRNSV. This planting is at least 1500' away from the nearest potential source of PNRSV in either peach or wild Prunus species.

At the present time 145 trees remain in the block. The losses represent an annual rate of re-infection of less than 1% together with some spread from initial foci of infection.

One variety, Tennessee Natural the seed of which is used to provide rootstocks, blooms at a later date than any other material in this planting. In 1984 a single tree in a row of 13 trees of Tennessee Natural was determined to be infected with PNRSV by using graft inoculation to Shiro-fugen flowering cherry. The infection was not detected until late summer. The tree was removed but in 1985 the remaining 12 trees were found to be infected with PNRSV and were eliminated. Assuming that the first tree identified to contain the virus was the initial focus of infection, we interpret this high rate of transmission in a specific variety to be due to a relatively few trees being "worked" by a large population of bees while there was no other flowering material available in the area at this time. In practice the considerable potential for the spread of this pollen borne virus within monocultures of peach varieties, once the initial focus of infection has been established, is probably reduced by the considerable number of trees that are available to a population of bees visiting an orchard.

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EFFECT OF WATER STRESS ON SUSCEPTIBILITY OF NONWOUNDED PEACH TREES TO *BOTRYOSPHAERIA DOTHIDEA*

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The disease known as peach gummosis caused by Botryosphaeria dothidea (Moug. ex. Fr.) Ces. & de Not. (=B. ribis Gross & Dugg.) was noticed in Georgia as early as the 1960's (Reilly et al. 1982, Weaver 1976) and has since spread to other areas of the Southeast (Pusey et al. 1986, Weaver 1976). Symptoms, as described by Weaver (Weaver 1974, Weaver 1979), are associated with lenticels. They include sunken necrotic lesions that frequently exude gum on trunks and scaffold limbs and swollen areas on young branches. All symptoms associated with lenticels have been reproduced in artificial inoculations of nonwounded peach trees with B. dothidea (Pusey et al. 1986, Weaver 1979), but not with other Botryosphaeria species found on peach (Pusey et al. 1986).

Prior to the work by Weaver (1974), Abiko and Kitajima (1970) gave a similar description for a peach disease in Japan first detected in 1965. Because of the characteristic swelling of bark tissue, it was called "peach blister canker". Gum was reported to ooze from the trunk and branches of trees. Diseased twigs eventually died. A fungus suspected to be the cause was shown through artificial inoculation to invade uninjured bark and induce blister formation. It was treated as a new species, Physalospora persicae Abiko et Kitajima. Koganezawa and Sakuma (1984) later suggested that the fungus be named Botryosphaeria berengeriana de Not. f. sp. persicae (Abiko et Kitajima) comb. nov. According to their study and a re-evaluation of Botryosphaeria by von Arx et al. (1975), B. berengeriana is a synonym of B.ribis,and the latter is not the same as B. dothidea as earlier established. They further indicated that most fungal isolates recently identified as B. dothidea by investigators should be B. berengeriana. It was specifically pointed out that the B. dothidea which causes peach gummosis in the United States should have been identified as B. berengeriana, based on a description of the fungus by Weaver (1974). Forma specialis persicae was proposed for the pathogen of peach blister canker in Japan (Koganezawa et al. 1984) because of its specificity to peach (Abiko et al. 1970). Likewise, B. berengeriana de Not. f. sp. piricola (Nose) comb. nov. was suggested in place of P. piricola Nose (Koganezawa et al. 1984), a pathogen specific to apple and the cause of "apple wart canker" in Japan. The finding of pathogenic types of B. berengeriana in Japan, incidentally, appears to correspond with work by Pusey et al. (1986) indicating that the B. dothidea in peach gummosis may be pathogenically unique from B. dothidea isolated from hosts other than peach. Concerning the argument for B. berengeriana instead of B. dothidea as used by some workers, until this gains acceptance, the author will use the latter name.

A gummosis disease of peach caused by B. dothidea was also recently reported by Chen (1985) to be a serious problem of peach production in Nanjing, China. The disease was considered by Chen to be the same as that described by Weaver (1974, 1979). Nonwound inoculations of trees in China led to blister formation and gumming associated with lenticels.

The reported time period between inoculation of nonwounded peach trees with conidia of B. dothidea (or B. berengeriana) and symptom expression varies from 2 wk to many months (Abiko et al. 1970, Chen 1985, Pusey et al. 1986, Weaver 1979). In one test, the author and coworkers (Pusey et al. 1986) did not observe symptoms in the same season that trees were inoculated, even though it was done in June. However, the following year, blisters, lesions, and gum exudation were observed. In another experiment, trees inoculated in August never expressed symptoms during 2 years of observation (Pusey, Reilly, and Okie; unpublished). This may be an indication that the time during the season when the fungus is introduced is important. There has also been some inconsistency in the type of symptom that develops when trees do show a visible reaction. Sometimes blisters appear, but lesion development and gumming never occur (Pusey, unpublished). B. dothidea has a reputation of being a stress pathogen (Crist et al. 1975, Weaver 1979); could it be that necrotic lesions develop more readily when plants are under stress? According to Connor (1968), B. dothidea can reside in lenticels of apple trees and may enter the cortex when moisture stress develops. The objective here was to study the effect of moisture stress on the response of nonwounded trees to B. dothidea.

MATERIALS AND METHODS

A B. dothidea isolate from diseased peach trees in Dooly Co., GA [designated as PI-5 in Pusey et al. (1986)] was used in all inoculations. The pathogen was transferred from Difco potato-dextrose maintenance medium to Difco oatmeal agar for production of conidia and grown under continuous light at 25 C for 21 days. Conidia were suspended in distilled water at 3.0 x 10^5 spores per milliliter. One-year-old budded Summergold peach trees were obtained from a nursery and placed in 9-inch plastic pots in early February, 1985. Four weeks later, the trees were fertilized with Osmocote 13-13-13. Water requirements were low initially because leaves were not fully expanded and outside temperatures were low. Trees were watered 5-6 days a week for 10 weeks. Sixteen days before making inoculations, trees were put on an automatic irrigation system which operated on an

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8-day cycle. Weighted tubes supplied water to individual pots. At 10 AM on the days that trees were watered, the pot was filled to capacity so that water drained from the bottom. Four different schedules were used. One group of trees received water every day, and three other groups had water withheld the last 2 days, the last 4 days, or the last 6 days of the 8-day cycle. Inoculations were made on May 10, which was at the end of the second 8-day cycle. Although some trees had not been watered for six days, there were no visible signs of water stress at the time of inoculation. The conidial suspension of B. dothidea was applied with a soft brush to a section of the tree stem extending from the bud union to the first branch (15-30 cm). Noninoculated check trees were brushed with water only. The stems were then wrapped with moist cheesecloth and Parafilm. Seven days later the wrappings were removed. For each of the four watering schedules there were 19 inoculated trees and 3 noninoculated trees. All trees were on one greenhouse bench in a complete randomized block design. The watering schedules were continued for 19 wk or 16 complete cycles. During this time the trees were examined periodically for symptoms. Data for the different treatments were compared using analysis of variance (P=0.05).

RESULTS

Trees never showed signs of stress, except during the period June 2-7 when outside temperatures exceeded 37 C. Five of the plants receiving the least amount of water were in a wilted condition which resulted in the loss of up to 25% of the leaves. Noninoculated check trees were free of disease symptoms throughout the experiment. Gum exudate was first observed on inoculated trees about 5 wk after inoculation. The total number of gumming sites on trees was determined June 18, July 3, August 27, and September 18. Until the last date, no gum was observed on trees watered every day (Fig. 1A), whereas some trees in each of the other groups showed gum on all dates. Trees deprived of water for 6 days in the 8-day cycle always had more gumming sites (P=0.05) per unit area of bark surface than other trees, except on the first two dates when no difference was shown between the above trees and those deprived of water for 4 days. Differences were never shown among the tree groups with water withholdings of 0, 2, or 4 days. On August 27, 72% of the trees with the greatest water deficit had gum. Gum exudate on a few trees dropped from the tree or dried up; thus, a decrease in percent of trees observed with gum was possible. Although this was shown for the latter group from August to September, those trees that continued to gum had an increased number of gumming sites (Fig. 1B).

On the final date, all gum deposits remaining on the tree were removed and weighed; and gum weight per unit area of inoculated bark calculated (Fig. 2A). Again, trees with the greatest water deficit were shown to be different (P=0.05) from those in the other treatment groups, which were not different from one another.

On the same day, numbers of blisters and necrotic lesions per unit area were determined from counts on one side of the tree (Fig. 2B). Blisters associated with lenticels had first been noticed about 13 wk after inoculation. Sites of gum production were also locations where lesions had developed, but not all lesions produced gum that was visible on the outside. There were no differences in the number of blisters per unit area of bark among the treatment groups, but the same statistical results given above for gumming sites and gum weight were shown for lesion number per unit area. Lesion diameters were used to calculate percent of the inoculated bark surface area that was necrotic. Although lesion areas ranged from 3 to 39 mm^2, percent of necrosis closely reflected lesion number per unit area when data were plotted (Figs. 2B and C) and statistical differences were the same. For trees with the greatest water deficit, 4.7% of the bark area was necrotic.

All trees were alive after 19 wk except one inoculated tree in the group with the greatest water deficit. The trees were transferred in September from the greenhouse to a lathhouse where they remained for a full year. There, each plant received essentially the same amount of water, either from rain or a sprinkler system. During the dry 1986 season they were occasionally neglected and may have been under some stress. All noninoculated trees were alive after one year in the lathhouse; however, 100% of the inoculated trees that received the least amount of water the year before were either dead or their inoculated stems had died and new shoots originated from the roots (Table 1). Percentages of trees in the other groups found in this condition ranged from 76 to 88%.

DISCUSSION

Water stress appears to affect the capacity of trees to resist infections at lenticels that lead to necrotic lesions and gum exudation. Stress is apparently not required since these symptoms were eventually expressed by a few of the trees watered every day. The mortality rates one year after the experiment are not very helpful in assessing the importance of stress in the observed death, because it is unknown whether or not the trees received a sufficient amount of water during the second season. Considering that none of the noninoculated stems died, it does point out that the fungus is capable of killing trees even when trees have not been injured prior to fungal invasions.

The formation of blisters associated with lenticels was not influenced by stress in the above experiment. Numerous blisters developed on inoculated trees in all groups, and means for the groups did not differ. The blisters consist largely of undifferentiated tissue beneath the lenticel and beneath the outer bark surrounding the lenticel. Their formation may be one mechanism of the plant to compartmentalize the fungus. Possibly lesions developed at some sites where blisters might have formed if plants had

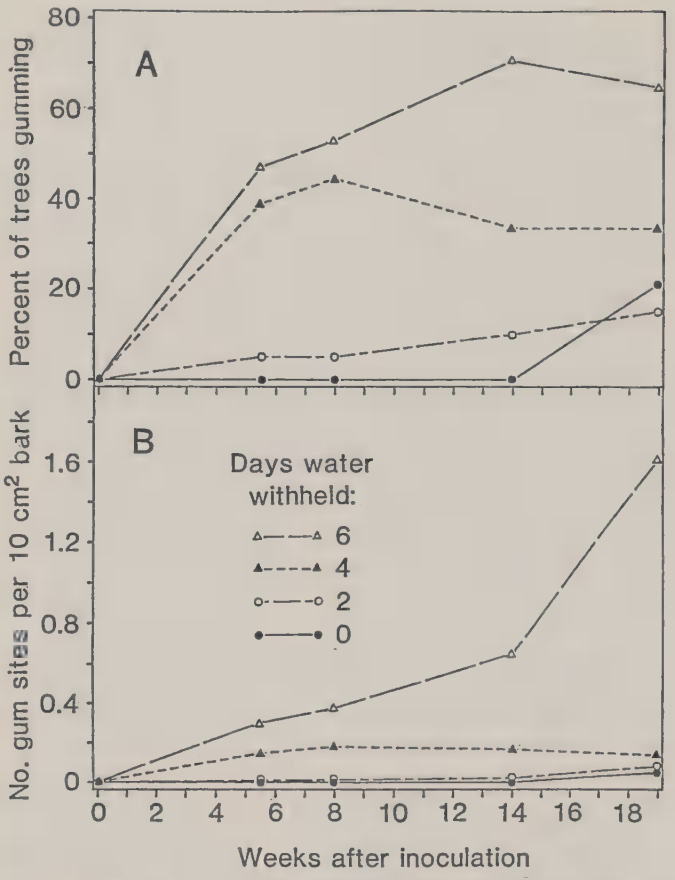

Fig. 1. Gumming response of nonwounded peach stems to Botryosphaeria dothidea during 19 wk subjection to water withholding for 0-6 days in an 8-day cycle. (A) Percent of trees exuding gum. (B) Number of gumming sites per 10 cm² of bark area.

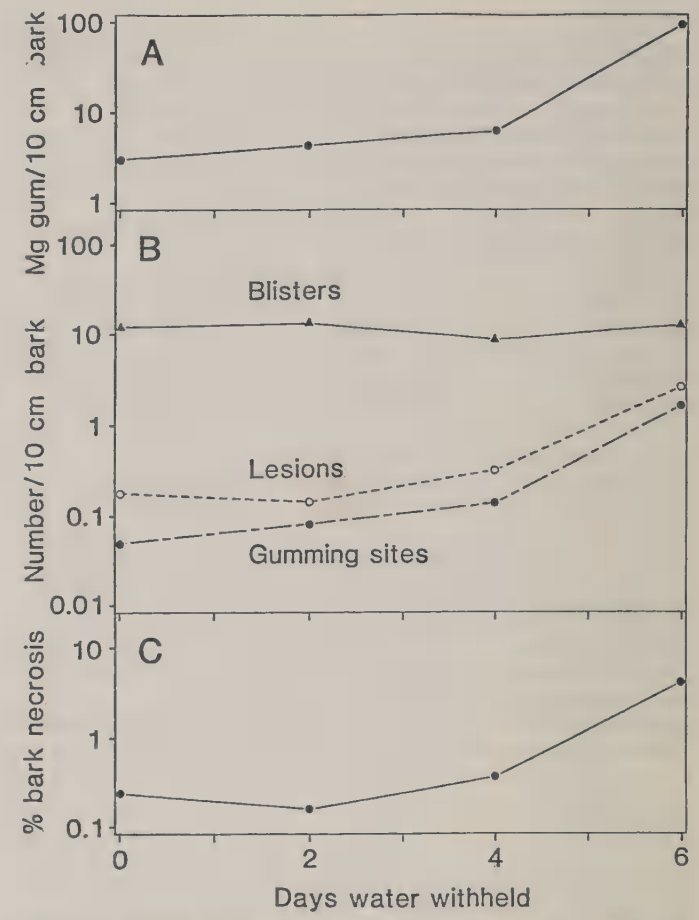

Fig. 2. Lenticel-associated symptoms of peach stems inoculated with Botryoshpaeria dothidea and subjected to water withholding for 0-6 days in an 8-day cycle. (A) Milligrams of gum exuded per 10 cm² of bark area. (B) Number of blisters, sunken lesions, and gumming sites per 10 cm² of bark area. (C) Percent of inoculated bark area that was necrotic.

Table I. Percent of trees inoculated with Botryosphaeria dothidea that were dead or had dead stems 1 year after a 19-wk subjection to water withholding for 0-6 days in an 8-day cycle.

Tree condition	Noninoculated check trees[1/]	Days water withheld from inoculated trees			
		0	2	4	6
Completely dead	0	23.5	5.9	29.4	41.2
Dead stem only[2/]	0	64.7	82.3	47.0	58.8
Completely dead or dead stem	0	88.2	88.2	76.4	100.0

1/ Check trees for all watering schedules combined.
2/ Tree dead above bud union but live shoots from root.

not been stressed. Occasionally, necrotic lesions were slightly raised rather than being sunken; so, either blisters can develop into lesions or an excessive amount of tissue is sometimes generated below developing lesions. According to Abiko and Kitajima (1970), blister formation on peach trees in Japan is generally followed by gum exudation from the blisters in later stages of the disease. This was also implied by Chen (1985).

Although water stress was shown to increase the susceptibility of uninjured peach bark to B. dothidea, the experiment did not tell us when it was a factor. Was it important throughout the 19-wk period or was it critical only soon after inoculation? Also, if trees had not been stressed until long after inoculation, what would the effect have been? Whether trees are stressed or not stressed, a question that may be of greater importance is: can the pathogen enter through lenticels and cause infection at any time during the year or only during a critical stage of lenticel development, and if the latter, when is this period? Hopefully, these and other questions can be answered in future investigations.

Realizing the importance of water stress in the development of this somewhat unique bark disease should be helpful in further attempts to study it. It should also be of value in developing an effective control strategy. Insuring that trees receive adequate water through the practice of irrigation would eliminate water stress as a predisposition factor.

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GENETICS AND ENVIRONMENT INFLUENCE THE FORMATION OF NEW TISSUES IN WOUNDED TREE BARK

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The proper management of wounds is critical for maintaining healthy, vigorous, and long-lived fruit orchards. Since wounds serve as infection courts for most types of pathogenic organisms in virtually all orchard crop systems, a better understanding of how trees respond to wounds can assist us in managing those systems where wound-diseases are economically important. A good example is Cytospora or Valsa canker of peach trees.

When a tree is wounded several nonspecific autonomous events are triggered depending upon the severity of the wound. Figure 1 was developed by Mullick (1977) and has been modified only slightly since it first appeared in the literature. The response to a wound which was limited to the phellogen and outer bark was characterized by a sequence of anatomical changes including 1) the formation of a primary ligno-suberized boundary zone, 2) the generation of a new phellogen immediately internal to the boundary zone, and 3) the formation of suberized phellem from the new phellogen. A slightly deeper wound disrupted normal functioning of the vascular cambium and resulted in the formation of altered cell types. This was followed by the three events described above. An even more severe wound resulted in the loss of functional sapwood through the compartmentalization process, followed again by formation of the boundary zone and new periderm. All three classes of wound severity had in common the formation of ligno-suberized tissue from cells present at the time of wounding and the generation of a new preformed barrier to pathogen ingress, the wound periderm.

Mullick hypothesized that the formation of the new periderm was the last event in a process that was triggered by wounding. He believed that trees responded to injuries, pathogens, or insects by attempting to generate new cambia where old cambia had been injured. Resistance to certain diseases and insects was a beneficial effect of this repair process. Thinking within this framework, Mullick stated that defense and repair were empirically inseparable and that the process of periderm generation was part of a nonspecific mechanism active in defense and triggered by wounding. This was a radical departure from the traditional view of wound periderm as a passive mechanical barrier, acting only to limit pathogen ingress beyond where it had been arrested by more active defense mechanisms.

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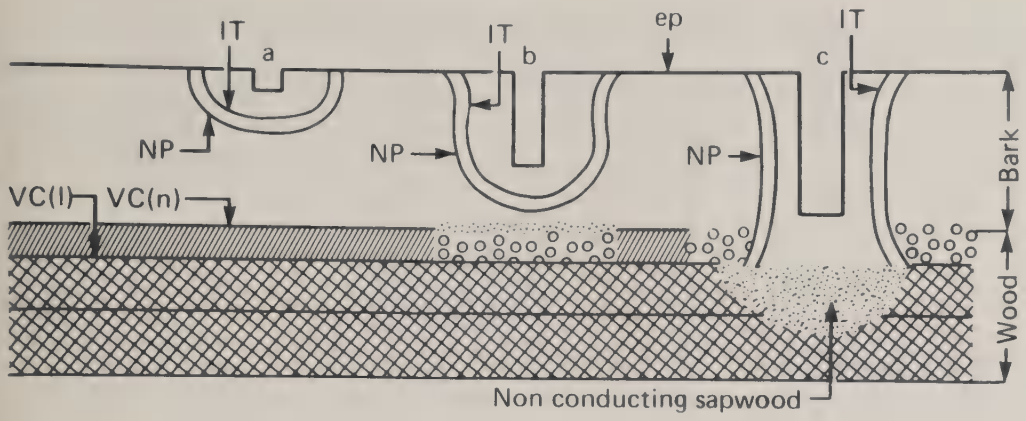
A critical aspect of the repair/defense process in trees is the influence of genetics and environment on the dynamics of meristem and tissue generation and the quantity and quality of new periderms. The generation rates for primary ligno-suberized boundary zones and new periderms, which could be interpreted as the rate of formation of new pre-formed barriers to pathogen ingress, are influenced by several environmental factors. Plant genotype also appears to influence both the rate of formation of new tissues and the amount of suberin in periderms of similar thickness.

The study of periderm regeneration is important because there are strong correlations between degree of wound healing and resistance to fungal infection. In a study to examine the influence of wound age on infection and colonization of peach bark by Leucostoma persoonii, we inoculated bark wounds of varying age with fungus mycelium (Biggs 1986b). Noninoculated wounds of similar ages and in close proximity to inoculated wounds were also sampled and examined histologically for morphological and histochemical changes associated with nonspecific plant defense reactions. This study revealed that lignified and ligno-suberized tissue were closely associated with a decreased rate of fungal colonization (Table 1). New periderm with at least 3 cells' thickness of suberized phellem was associated with immunity from fungal colonization within the time frame of the experiment (Table 2). The critical period regarding effective periderm formation was between 10 and 14 days post-wounding. Many other studies using different host/pathogen combinations have shown a similar relationship between wound response and resistance to infection. It is reasonable to assume that wounds on peach trees could require 10-14 days of protection to allow time for the tree's natural defenses to impart disease resistance. Middleton and Bostock have reached a similar conclusion in their work with Ceratocystis canker of almond in California (Middleton and Bostock 1985).

This 10 to 14 day protection interval obviously is not the generalized wound response time of a tree grown under all conditions present in a typical orchard. There are several host, pathogen, and environmental factors which influence the time required for a new and effective periderm to form.

In experiments to determine how quickly periderm forms when wounds are made at intervals during the season, we were able to show that depending upon when the wounds were inflicted, the number of days required for periderm to form varied. For example, wounds on peach made in early May required about 21 days to form periderm compared to 10 days for wounds made in mid July. From earlier studies on wound response in leaf scars we knew that temperature greatly influenced the formation of the boundary zone and new tissues (Biggs and Northover 1985).



NP – Necrophylactic periderm
ep – Exophylactic periderm
VC(n) – Position of vascular cambium at time of phellogen restoration
VC(I) – Position of vascular cambium at time of injury
– Conducting sapwood formed after injury
– Conducting sapwood extant at time of injury
– Transformed cambia-phloic zone
– Zone of newly restored VC
– Zone of non-conducting sapwood

(Mullick 1977 – Modified)

Fig. 1. Illustration of woody plant response following wounding to different depths, modified after Mullick (1977). Wounds are (a) shallow and limited to the active phellogen and bark cortex, (b) deeper and limited to the inner bark without mechanical damage to the vascular cambium, or (c) through the bark and vascular cambium to the xylem surface. All three wounds have in common the formation of a ligno-suberized boundary zone (IT) and necrophylactic (wound) periderm (NP).

TABLE 1. Canker length (mm) and linear regression coefficients for the relationship between canker length (Y) and time (days, X) following inoculation of peach bark wounds of varying age with mycelium of Cytospora leucostoma

Wound age (days)	Wounds infected (%)	Canker length (+ 6.0 mm) Time postinoculation (days) 3	7	14	21	Regression coefficient
CK	0	6.0 [y]	6.0	6.0	6.0	0.388a[z]
24	0	6.2	6.0	6.0	6.2	0.395a
14	10	6.6	9.2	12.2	15.4	0.832a
10	100	6.6	13.0	18.6	24.6	1.277 b
7	100	9.2	18.4	26.6	27.4	1.589 b
3	100	14.2	20.2	26.2	30.2	1.705 bc
0	100	20.2	27.2	39.2	49.8	2.656 c

[y] Data are means of measurements from 10 trees from one experiment.
[z] Analysis of covariance for testing homogeneity of regression coefficients was significant, F = 9.31 (P = 0.01). Letters denote significantly different regression coefficients using paired t-tests in all combinations.

TABLE 2. Summary of morphological and histochemical changes related to phellogen generation following wounding in peach bark

Wound Age	Lignified boundary[w]	Ligno-suberized boundary[w]	Necrophylactic periderm[x]	Suberized xylem ray parenchyma[w]	% transmission (lignin)[y]	Autofluorescence intensity (suberin)[z]
0	-	-	0	-	100	0.0
3	+	-	0	-	97.3	0.0
7	+	+	0	+	76.7	3.1
10	+	+	1.0	+	66.0	15.2
14	+	+	3.0	+	67.1	21.2
24	+	+	6.0	+	61.4	33.3

[w] feature present (+) or absent (-) in the outer bark cortex in contact with the original periderm in 100% of plants examined. For suberized xylem ray parenchyma, signs indicate presence or absence in 100% of plants examined.

[x] Mean number cells counted in transverse section at the junction of the new and the original periderms (n = 20).

[y] Percent transmission at 546 nm measured over a circular area with 272 um diameter, each measured area contained about 100 cells. Values represent mean percent transmission (n=20) of ligno-suberized tissue or ligno-suberized tissue plus necrophylactic phellem on wound age. Nonwounded bark percent transmision = 100.

[z] Autofluorescence intensity measured over a circular area with 272 um diameter, each measured area contained about 100 cells. Values represent mean autofluorescence intensity (n = 20) of ligno-suberized tissue or ligno-suberized tissue plus necrophylactic phellem depending on wound age. Nonwounded bark autofluorescence intensity = 0.

At each postwounding time at each wound date (seven for peach), data were compiled on the percentage of wounds lignified and suberized. Mean daily temperatures were monitored, and the accumulated degree days using bases of -5°, 0°, 5°, and 10° C were calculated for each wound series and sampling time. Regression analyses were conducted to investigate the relationship between wound responses (lignification and suberization) and time (days) postwounding, mean daily temperature during the postwounding period, and accumulated degree days. Regression coefficients were tested for homogeneity using analysis of covariance and accumulated degree days (base = 0°C) was the best predictor of wound response in these analyses. When the seven linear regressions calculated for the seven individual wound series were compared, analysis of covariance showed that the slopes of the seven lines were homogeneous. This suggested that tree developmental stage during the period of May through August had little influence on wound healing rate. When these data were pooled and subjected to analysis using a segmented quadratic equation, the following equations were derived:

$$Y = -23.45 + 0.37x + 0.00036x^2,$$

where Y = Percent of wounds lignified, x = accumulated degree days and $R^2 = 0.86$; and

$$Y = -28.67 + 0.25x + 0.00015x^2,$$

where Y = Percent of wounds suberized, x = accumulated degree days, and $R^2 = 0.80$ (Fig. 2). For peach 256 degree days are needed for complete lignification and 411 degree days are needed to complete suberization. Sweet cherry and apple formed new tissues faster than peach. These data suggest that an important relationship exists for temperature and wound response and demonstrate the presence of species differences in the parameters examined (Biggs 1986a).

Using historical weather data on accumulation of degree days averaged over the past 40 years, the number of days from different pruning dates for wounds to lignify and suberize can be estimated. For peaches pruned on March 15 in Ontario, it would take 49 and 62 days, respectively, for the boundary zone and periderm to form completely around the wound. When pruned on May 15, these events would take place in 18 and 27 days, respectively. These data (from wounds on five-year-old Redhaven tree trunks) show that protection intervals longer than 10 to 14 days may be required to protect peach from infection by Leucostoma species, particularly when trees are pruned early.

Plant water status is also known to influence the susceptibility of trees to fungal pathogens (Schoeneweiss 1981). In experiments to determine the influence of irrigation on wound response in peach bark, we were able to show with two years of data, that irrigated trees form suberized wound periderm more quickly than nonirrigated trees (Table 3), and Bob Cline has been able to show that irrigated trees are less cankered than nonirrigated trees. The difference between treatments was shown to be due to differences in the rate of formation of suberized phellem cells in the new periderm (Biggs and Cline 1987).

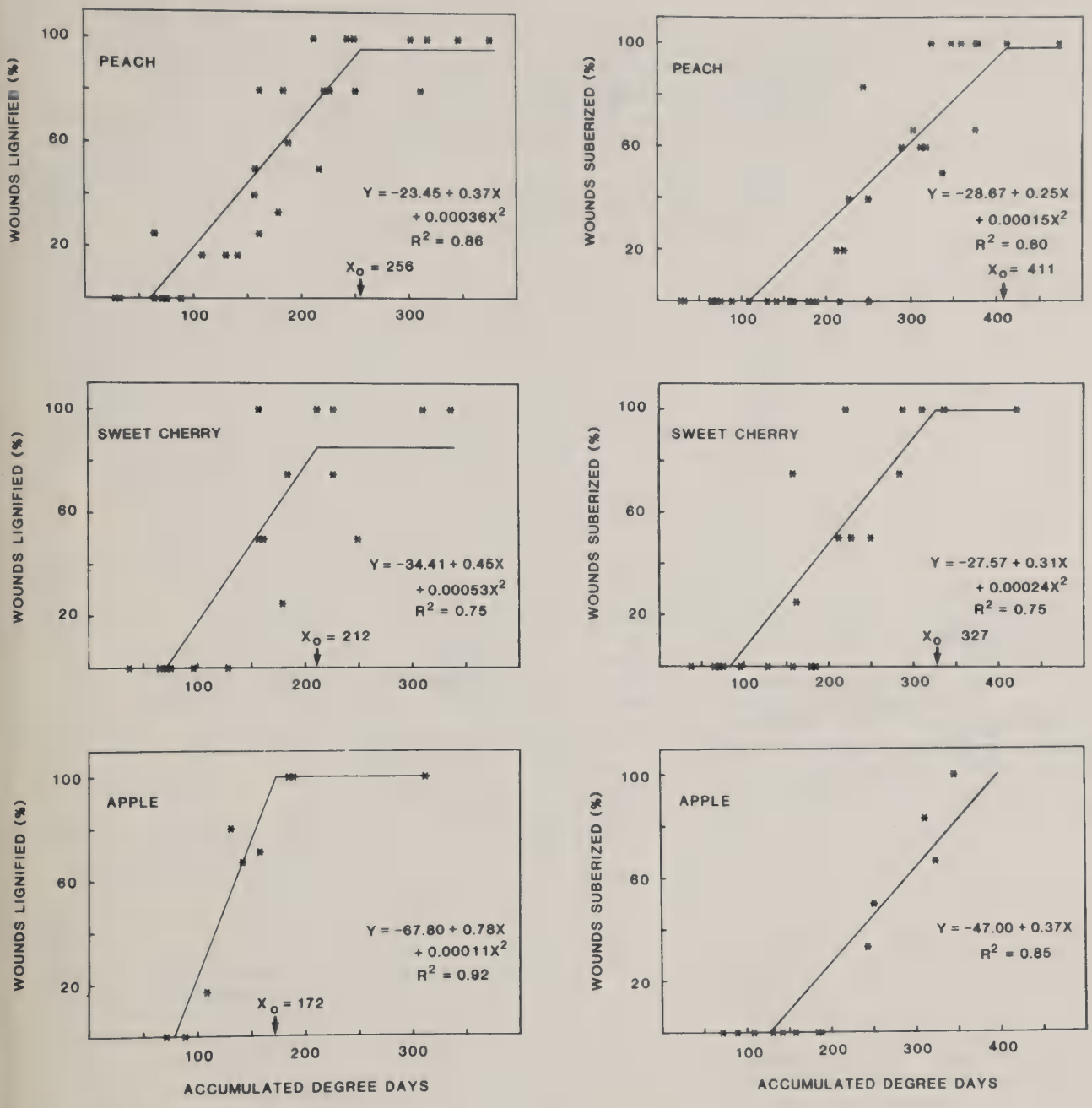


Fig. 2. Segmented quadratic and linear models (% wounds suberized in apple) for the relationship between two wound responses (lignification and suberization) and accumulated degree days following wounding (base = 0°C). Each asterisk represents 4 to 6 observations.

TABLE 3. Influence of irrigation on suberin autofluorescence intensity in the necrophylactic periderm of wounded bark of the peach cultivar Candor.

Wound date and days postwounding	Autofluorescence intensity, mV	
	Nonirrigated	Irrigated
22 May 84 (irrigation off)		
10	0.64	0.71
14	2.26	2.56
17	7.23 (0.257)a	8.48 (0.299)a
26 June 84 (irrigation off)		
10	2.84	3.15
14	7.56	9.34
17	10.53 (0.514)a	11.14 (0.530)a
24 July 84 (irrigation on)		
10	1.16	0.80
14	5.14	5.92
17	9.60 (0.390)a	11.05 (0.428)b
14 June 85 (irrigation on)		
10	0.10	0.64
14	1.24	4.30
17	5.24 (0.173)a	11.52 (0.414)b

[z] Autofluorescence intensity measured over a circular area 272 um diameter; each measured area contained about 100 cells. Values represent suberin autofluorescence intensity of primary ligno-suberized boundary zone (impervious tissue) or boundary zone plus necrophylactic phellem, depending on wound age. Nonwounded bark autofluorescence intensity = 0. Values in parentheses are slopes of regression lines for rate of suberin autofluorescence accumulation over seventeen days. Each value represents the pooled data from 8 replicate trees and 3 measurements per tree. Different letters in rows denote significant differences in regression line slopes determined by analysis of covariance and the F-test (P = 0.01).

Genetic differences within a single species has an effect on the quality of the wound periderm in peach. In our current research to assess wound response as a factor correlated with disease resistance in peach, we have observed dramatic differences between clones in the amount of suberin in the wound periderm, measured fluorometrically as autofluorescence intensity. These data suggest differing genetic capacity to synthesize and structurally incorporate the suberin polymer into the cell walls of the new periderm. The quantity of suberin appeared strongly correlated with the performance history of these clones as regards the peach canker disease (Table 4). However, this relationship must be interpreted with caution since it was repeatable in May and June and was absent in experiments conducted in other months.

To summarize, we have presented data which demonstrated how genetics and environment influenced the ability of trees to form new tissues after wounding. Protection of wounds for at least 14 days in a manner that does not inflence negatively the natural plant repair mechanism is probably essential to reduce the impact of wound-associated diseases. This is currently under investigation in our laboratory. It is important to realize that the wound repair process is influenced by temperature and soil moisture and that wound repair time will vary depending upon when wounds are inflicted. Cultivars also vary in their capacity to form periderms. This quality may be useful in identifying peach clones less susceptible to Cytospora canker. Many other host, pathogen, and environmental variables may influence events following wounding. Host carbohydrate reserves, the presence of the pathogen and its inoculum density, and site factors, disease history, foliar pathogens, and bark epiphytes could all affect wound response.

Further studies are required to clearly define the importance of these variables.

Table 4. Relationship between suberin deposition rate (b, millivolts autofluorescence intensity per day) in bark following wounding and field performance history (field rank) (1 = least susceptible, 10 = most susceptible) of 10 peach cultivars (May 1986).

Cultivar	b	b rank	field rank
V68101	.332	1	1
Veeglo	.299	2	3
Babygold	.249	3	4
V68051	.244	4	2
Vanity	.237	5	5
Redhaven	.227	6	6
Candor	.217	7	7
Madison	.203	8	8
Earlired	.166	9	10
Vivid	.114	10	9

Spearman's rank correlation, $r_s = 0.952$, $t = 8.79$, $P \leq 0.01$.

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PEACH TREE SHORT LIFE IN PERSPECTIVES OF GAS EXCHANGE ACTIVITIES.

Umedi L. Yadava[1]

INTRODUCTION

Marginal production efficiency and longevity of peach trees have resulted from certain problems related to fruit growing such as peach tree short life (PTSL) syndrome. This problem of PTSL is of great concern especially in the Southeastern area of the United States, which produces about 35% of the nation's annual peach crop. Typically, tree death due to PTSL involves sudden collapse of new growth following spring budbreak. In most cases, tree mortality is preceded by either or both of winter/cold injury as manifested by trunk cambial browning (TCB) and bacterial canker (Pseudomonas syringae pv. syringae van Hall) damages (Yadava and Doud 1978, 1980). Plant growth substances or phytohormones which regulate physiodormancy and influence cold/winter hardiness of plant tissue, have also been implicated in the alteration of leaf gas exchanges (Bradford 1983, Cornic et al. 1983, Kriedemann et al. 1975, Martin et al. 1981, Ryc and Lewak 1982, Yadava et al. 1986). The transition in tree physiology from the state of normal health to that of PTSL development or similar stress conditions may be in some way associated with **invisible** or **undetected injuries** and **subtle** or **metabolic changes.** These kinds of deviations in plant health, especially in relation to various disorders, have been indicated in the literature (Basiouny and Biggs 1976, Cornic et al. 1983, Frenyo and Buban 1976, Gottwald and Wood 1985, Martin et al. 1981, Tan and Buttery 1982). However, in order to understand the mechanism of PTSL development, these subtleties and invisible changes have not been investigated so extensively using such basic physiological tools as certain gas exchange activities (GEA) to clearly explain the devastating PTSL syndrome of peach trees. Furthermore, it appears that the nature of the so called sudden collapse of PTSL affected trees may not be as sudden as it has been generally considered thus far. Therefore, in view of this consideration, the basic knowledge of PTSL syndrome in relation to GEA and their modification by PTSL causing and/or predisposing factors may increase the possibility for finding practical solutions to effectively manage the peach tree short life syndrome.

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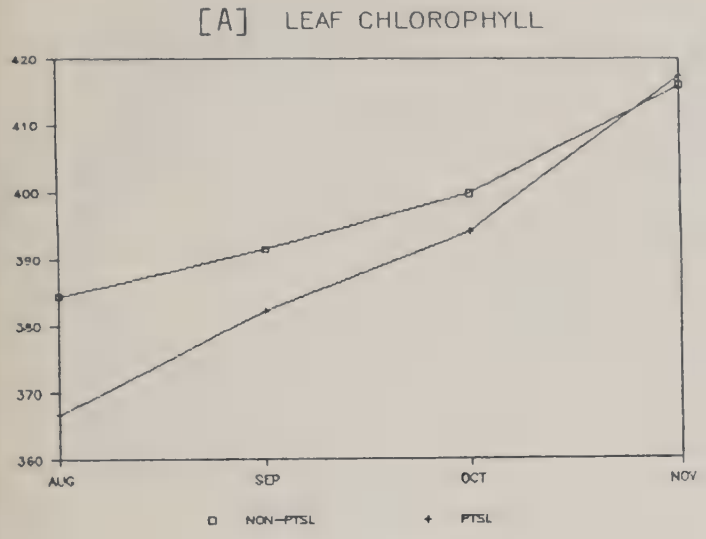
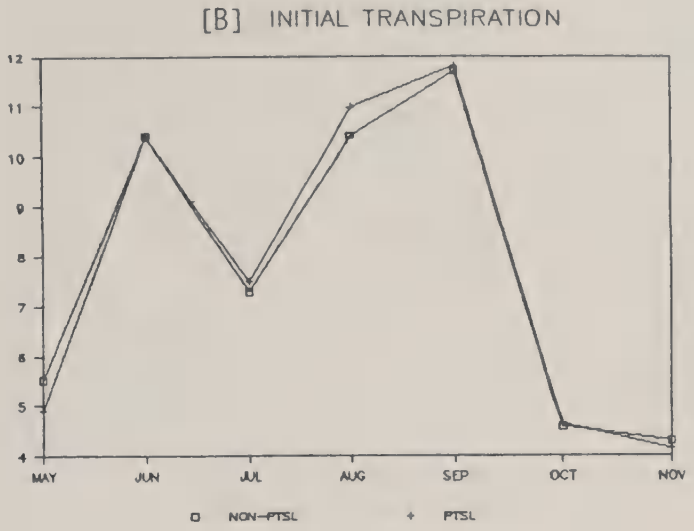
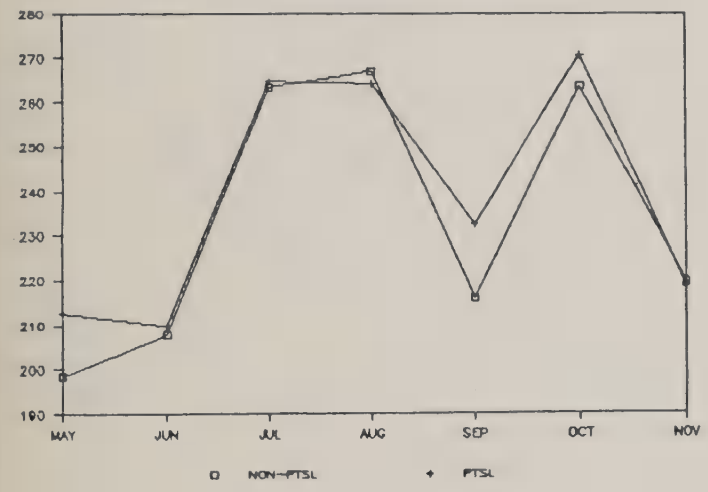
MATERIALS AND METHODS

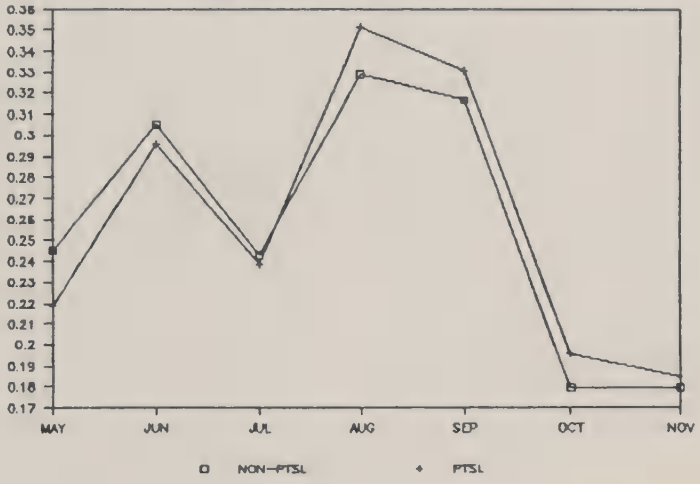
Three experimental plantings were used in this investigation. (1) A 6-year-old planting of 'Redhaven' peach (Prunus persica (L.) Batsch) trees grafted on 'Lovell' and 'Siberian C' peach seedling rootstocks (RSTK). Experimental trees in this planting were sprayed 3 times annually from 1981 to 1985 with 100 $\mu g\ L^{-1}$ each of abscisic acid (ABA), benzylaminopurine (BAP), gibberellic acid (GA_3), indoleacetic acid (IAA), and a mixture of ABA, BAP, GA3 and IAA. Distilled H_2O spray was used as a control. There were four replications for each treatment combination. (2) Second-leaf trees of 'Redhaven' on 'Lovell' and 'Nemaguard' peach seedling RSTK growing in the microplots of 55-gallon barrels filled with soil from PTSL and non-PTSL sites. All trees were sprayed with 100 $\mu g/L$ ABA or GA_3, and distilled H_2O was used as a control. All treatment were replicated 3 times. (3) A 4-year-old planting consisting of peach seedling trees of domestic 'Lovell' and 'Wild peach' from the Himalayan foothills, which were planted in two rows with six replications. Due to very small leaf size as well as greater than 60% mortality of the seedling trees of 'Amaru' and 'Baim', these two Himalayan germplasm were not included in the present investigation.

Two trees per treatment combination were selected for measuring GEA data throughout the 1985 growing season. Fully expanded leaves between the ages of 15 and 20 days were selected in late April on the southern half (sunny side) of tree canopy about 5 feet above ground on trees in experiments 1 and 3 but only 3 feet above ground in experiment 2. Gas exchanges, were repeatedly monitored each month on the same individual tree leaves, from early May to middle of November 1985. The gas exchanges data were collected for initial rate of transpiration (MTR), initial cellular or internal concentration (concn) of CO_2 (IC2), relative humidity (MRH), leaf and chamber temperatures (MLT, MCT), light intensity (MQU), ambient CO_2 concn (MC2), range of CO_2 consumption by leaf surface (10 cm^2) exposed in the chamber (RC2), and the rate of gaseous flow through leaf chamber (MFL), stomatal conductance (MCM), and net photosynthesis (MPM). A portable photosynthesis system (LI-6000) was employed to collect these data from 0900 to 1530 hour whenever an adequate light intensity (>800 $\mu E\ m^{-2}\ s^{-1}$) was available in the orchards. For data on the area concentrations of chlorophyll (CHL) of the same leaves which were used for GEA data collection, readings were taken on an electronic chlorophyll meter (SPAD-501, Minolta Corporation) prior to GEA data as described by Yadava (1986). Selected data from LI-6000 observations along with CHL data from SPAD meter, were directly transferred to an IBM/PC and then to the mainframe computer. Area concn of leaf chlorophyll ($\mu mol\ m^{-2}$) were calculated from SPAD-501 readings using a straight line formula ($Y = 9.120822X - 11.56395439$) recently developed by Yadava at the Agricultural Research Station of the Fort Valley State College. All data were analyzed using analysis of variance, F-test of significance and correlation coefficients. The Graphics partner and Lotus software programs were used to create graphics presented in figures 1 through 6.









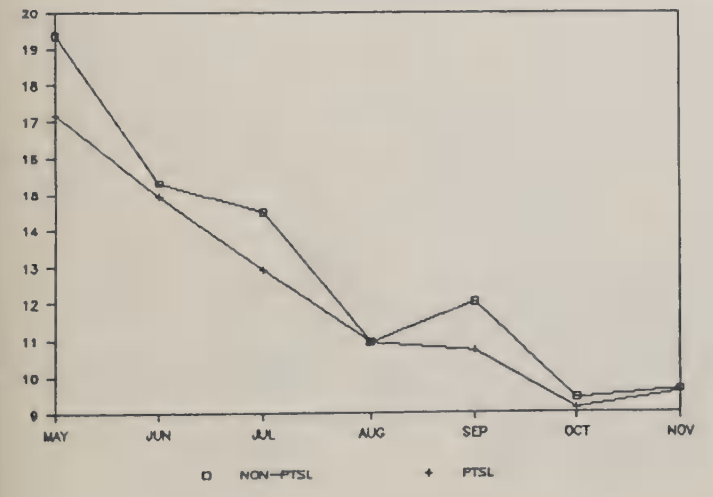
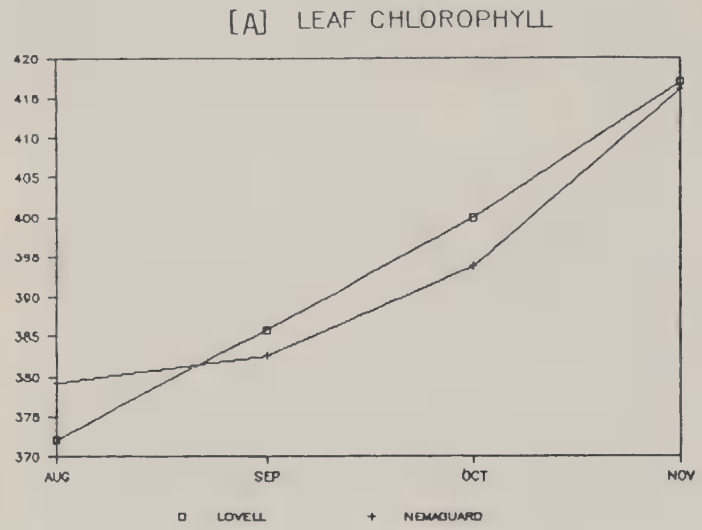
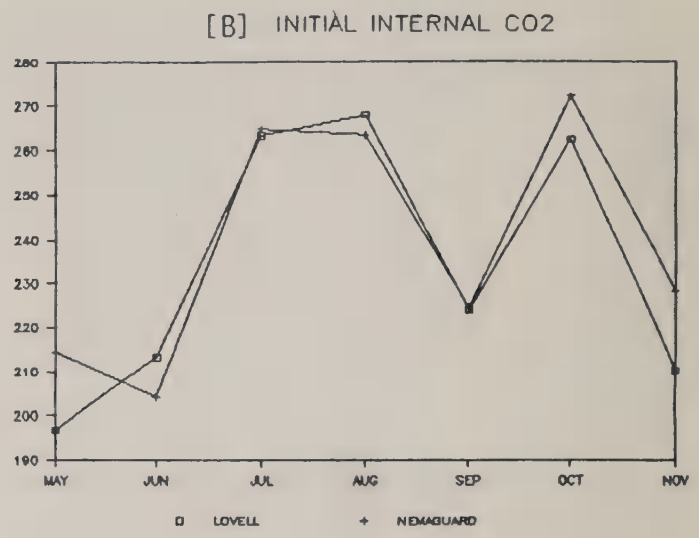
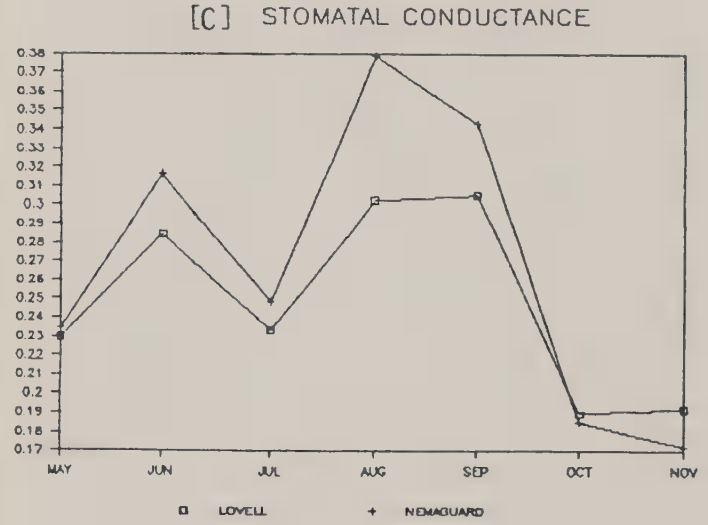
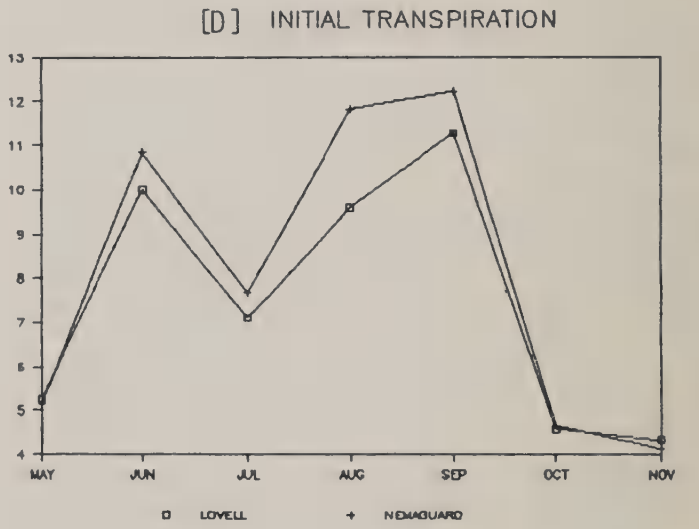


Figure 1
Influence of soil type (in microplots) on leaf chlorophyll concn [μ mol m^{-2}], rate of initial transpiration [μ mol H_2O m^{-2} s^{-1}], initial internal CO_2 concn [μl l^{-1}, stomatal conductance [mol m^{-2} s^{-1}], and rate of photosynthesis [μmol CO_2 m^{-2} s^{-1}] during 1985 growing season.









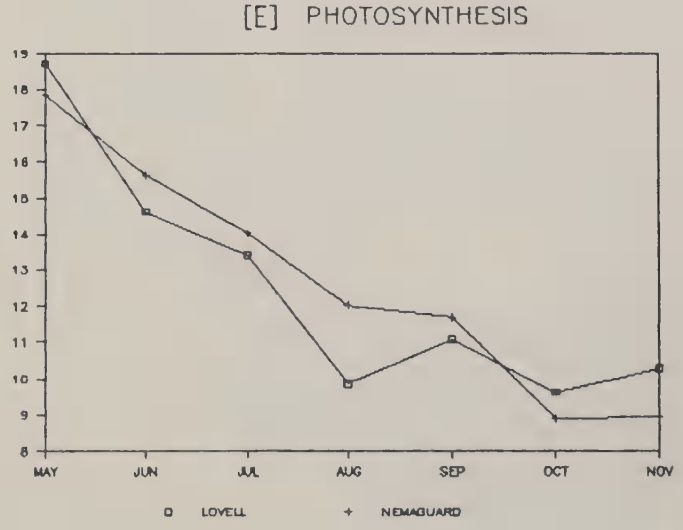
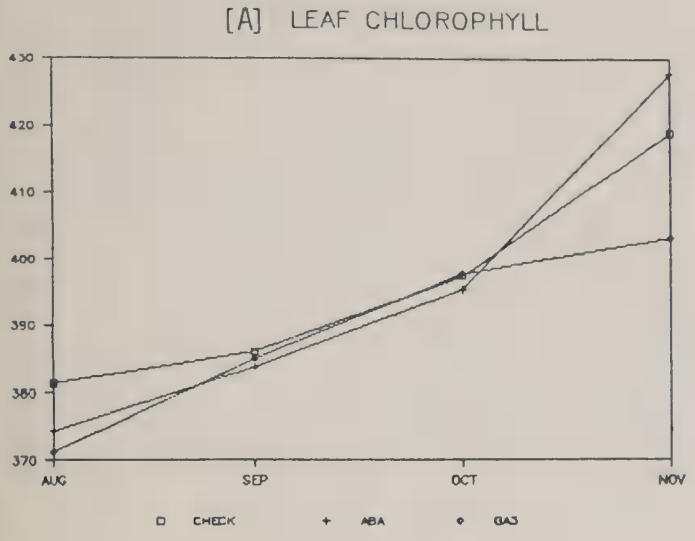
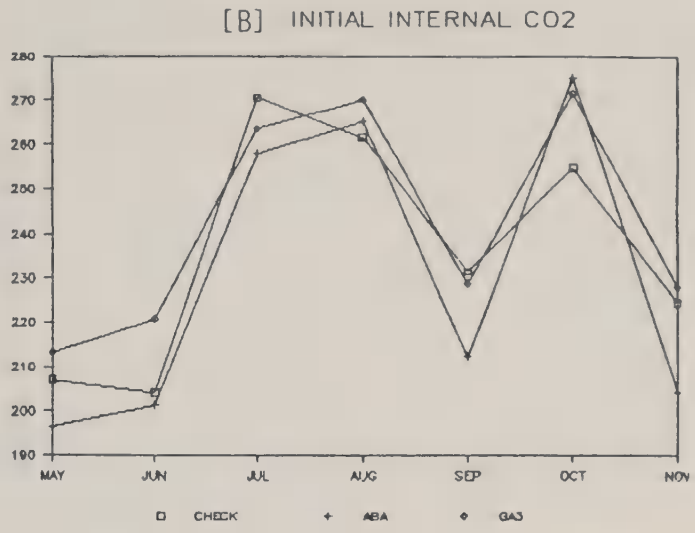
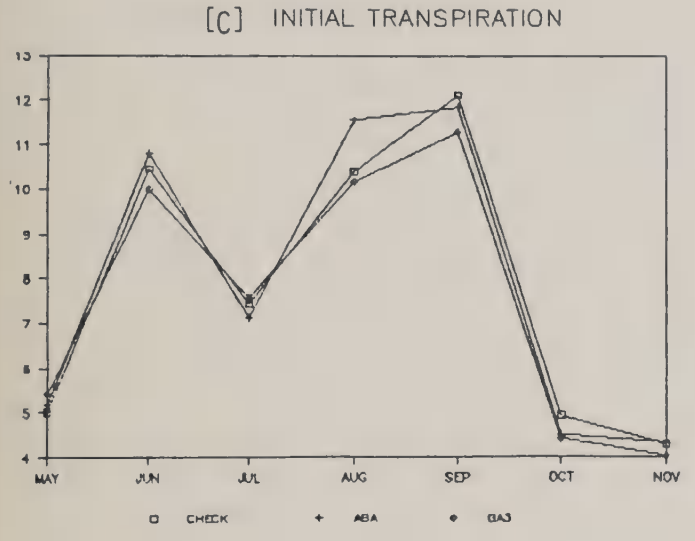
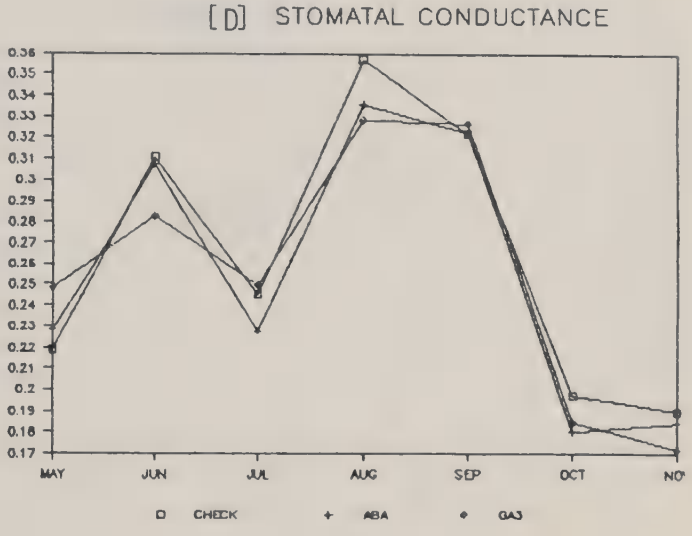


Figure 2
Influence of rootstock (in microplots) on leaf chlorophyll concn [μmol m^{-2}], rate of initial transpiration [μmol H_2O m^{-2} s^{-1}], initial internal CO_2 concn [μl l^{-1}, stomatal conductance [mol m^{-2} s^{-1}], and rate of photosynthesis [μmol CO_2 m^{-2} s^{-1}] during 1985 growing season.









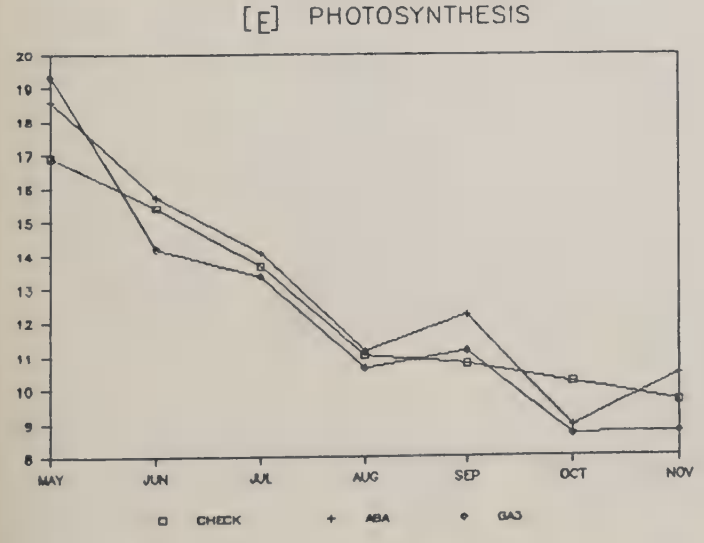


Figure 3
Influence of phytohormone (in microplots) on leaf chlorophyll concn [μ mol m^{-2}], rate of initial transpiration [μ mol H_2O m^{-2} s^{-1}], initial internal CO_2 concn [μl l^{-1}, stomatal conductance [mol m^{-2} s^{-1}], and rate of photosynthesis [μ mol CO_2 m^{-2} s^{-1}] during 1985 growing season.

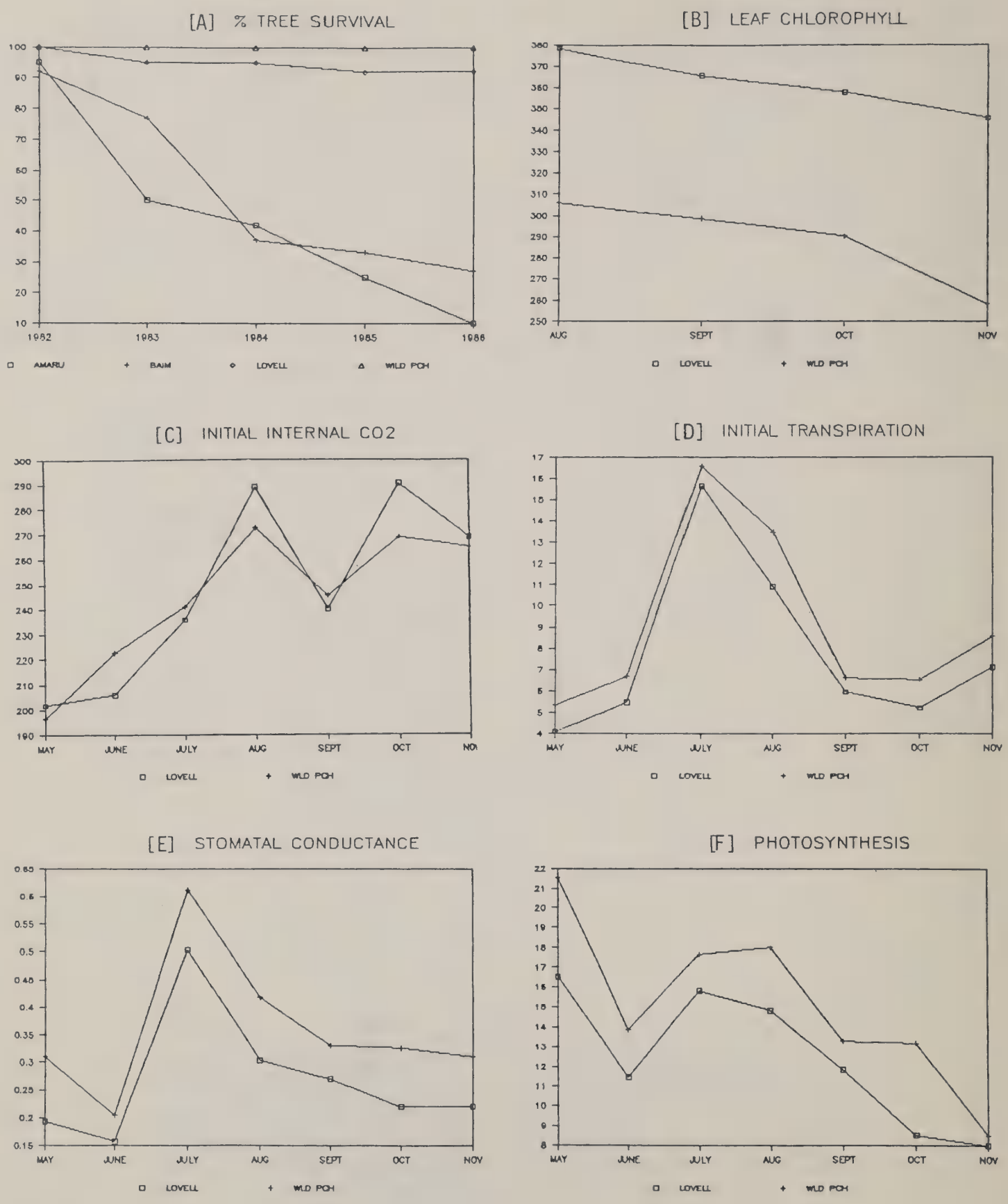


Figure 4
Influence of peach germplasm on tree survival (1982-1986) and leaf chlorophyll concn [µmol m^{-2}], rate of initial transpiration [µmol H_2O m^{-2} s^{-1}], initial internal CO_2 concn [µl l^{-1}], stomatal conductance [mol m^{-2} s^{-1}], and rate of photosynthesis [µmol CO_2 m^{-2} s^{-1}] during 1985 growing season.

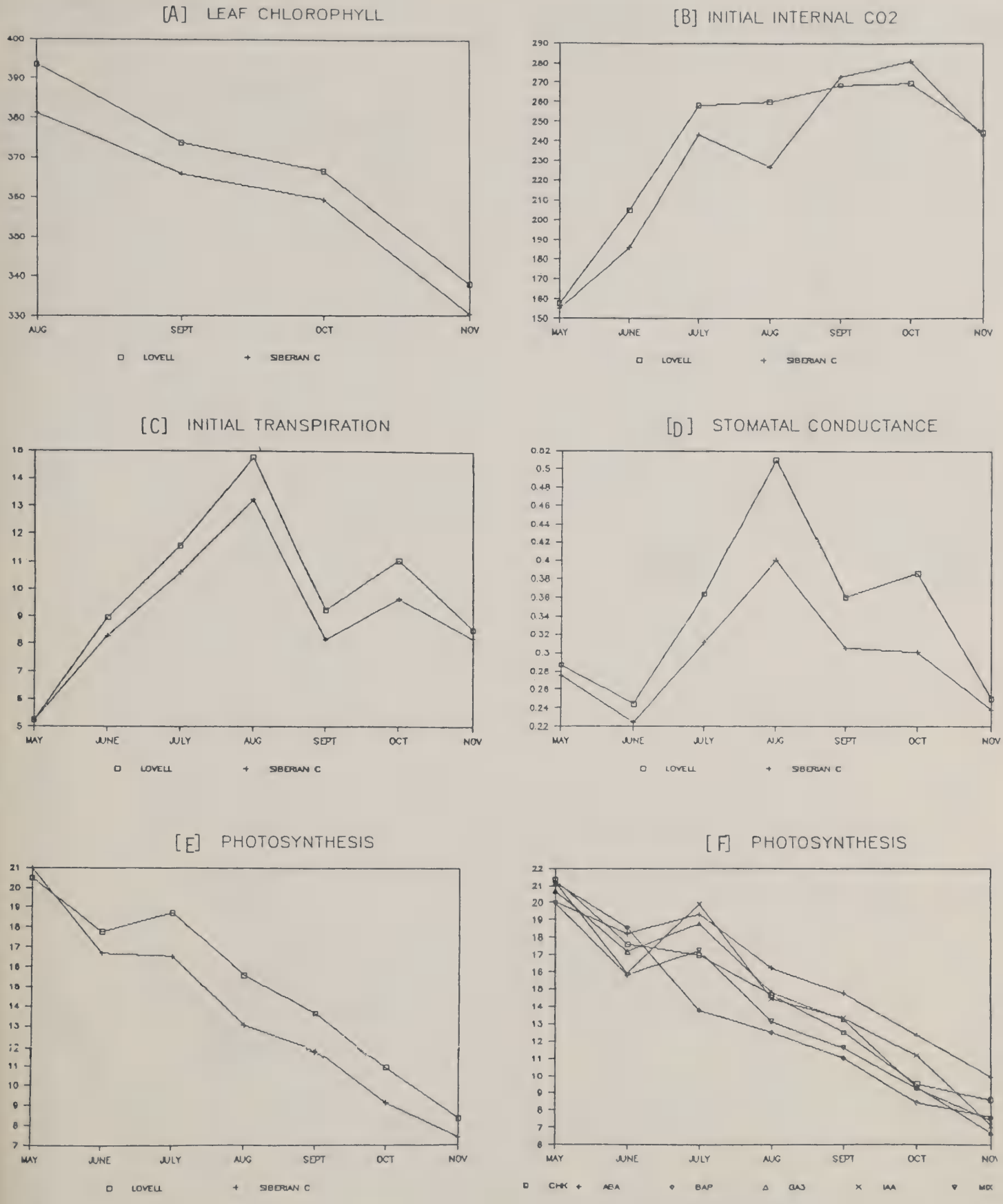


Figure 5
Rootstock influence on leaf chlorophyll conc [μ mol m^{-2}], rate of initial transpiration [μ mol H_2O m^{-2} s^{-1}], initial internal or cellular CO_2 conc [μl l^{-1}], stomatal conductance [mol m^{-2} s^{-1}], and rate of photosynthesis [μmol CO_2 m^{-2} s^{-1}], and phytohormone effect on rate of photosynthesis [μmol CO_2 m^{-2} s^{-1}] during 1985 growing season.

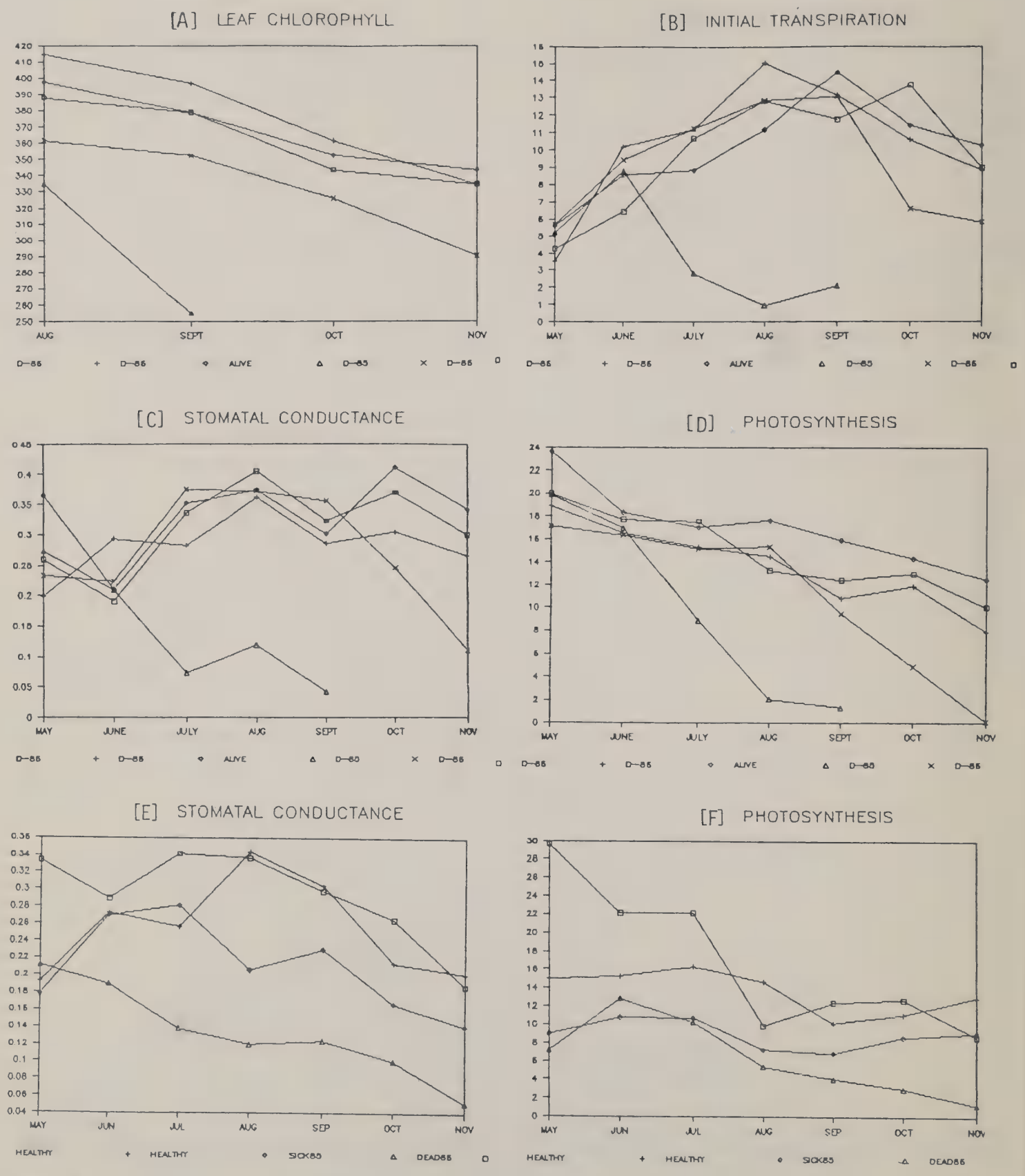


Figure 6
Response of Redhaven peach trees (ranging from normal to marginal health) for leaf chlorophyll concentrations [μmol m^{-2}], initial transpiration rate [μ mol H_2O m^{-2} s^{-1}], stomatal conductance [mol m^{-2} s^{-1}], and photosynthesis [μ mol CO_2 m^{-2} s^{-1}] in a field planting, and for stomatal conductance [mol m^{-2} s^{-1}] and rate of photosynthesis [μ mol CO_2 m^{-2} s^{-1}] in microplots during 1985 growing season.

RESULTS AND DISCUSSION

GENERAL:

In these investigations, the single leaf GEA of field-grown peach trees as expressed on leaf area basis showed that the rate of photosynthesis of these leaves was significantly correlated with the rates of transpiration and stomatal conductance, cellular CO_2, and area concns of CHL (Table 1). These kinds of correlations for such parameters have been reported in the literature by several researchers (Basiouny and Biggs 1976, Buttery and Buzzell 1977, Cornic et al. 1983, De Jong 1982, Kriedemann et al. 1975, Rom and Ferree 1985, Tan and Buttery 1982, Zelitch and Waggoner 1962). The negative correlation of IC2 with MPM in most cases in the present study, has been reported by De Jong (1982, 1983), but Cornic et al. (1983) found a direct relationship between the partial pressure of initial CO_2 and MPM. The average value of initial cellular CO_2 concn for these experiments was 241.19 $\mu l\ L^{-1}$. This value for peach, as a C3 plant species, is typically within the range of 230 to 260 $\mu l\ L^{-1}$ internal CO_2 concn when ambient CO_2 concn is around 320 $\mu l\ L^{-1}$ provided that the water, light, and temperature conditions were not the limiting factors (Farquhar et al. 1980).

The average MPM rate for all types of peach trees (young and old, vigorous and healthy, in marginal health, and unhealthy) during the 1985 growing season was 13.52 $\mu mol\ CO_2\ m^{-2}\ s^{-1}$. These rates of single leaf MPM for peach leaves are in the range of and conform with the findings reported by other workers (Basiouny and Biggs 1976, Chalmers et al. 1975, Cornic et al. 1983, Crews et al. 1975, Tan and Buttery 1982, and De Jong 1982). In general, trees with visible or invisible stress conditions or those trees growing on soil from the PTSL site, had lower MPM than their counterparts (Figs. 1-E, 6-D, 6-F). This may have resulted from certain type of changes in trees physiology imposed by the various kinds of stresses (Basiouny and Biggs 1976, Chalmers et al. 1975, Cornic et al. 1983, Gottwald and Wood 1985, Kriedemann et al. 1975, Martin et al. 1981, Ryc and Lewak 1982, Tan and Buttery 1982, Zelitch and Waggoner 1962). Thomas (1955) indicated that in a complicated system of plant-environment interaction, several important factors such as ambient air temperature, leaf age, soil moisture and nutrient levels, phytotoxins, and plant growth inhibitors influence MPM rate indirectly through changing the leaf morphology, and/or by modifying the enzyme systems and plant health. A proportional reduction in the MPM rate of pecan foliage to the amount of surface area of canopy colonized by pecan scab -causing organism

Table 1: Pearson correlation coefficients between various GEA parameters.@

	MTR	IC2	RC2	MCM	MPM
EXPERIMENT # 1 [N = 611, 374 for CHL]					
IC2	0.224*				
RC2	0.143*	-.270*			
MCM	0.811**	0.181*	0.288*		
MPM	0.156*	-.288*	0.976**	0.295*	
CHL	0.363*	-.039	0.500*	0.406*	0.467*
EXPERIMENT # 2 [N = 460, 177 for CHL]					
IC2	-.031				
RC2	0.128	-.306*			
MCM	0.744**	0.013	0.387*		
MPM	0.137*	-.393*	0.839**	0.479**	
CHL	-.243*	-.432*	0.033	-.100	0.330*
EXPERIMENT # 3 [N = 168, 72 for CHL]					
IC2	0.158*				
RC2	0.228*	-.291*			
MCM	0.848**	0.141	0.412*		
MPM	0.290*	-.293*	0.965**	0.459**	
CHL	-.046	0.093	0.144	-.263*	0.141

* Significant at <0.05 level
** Significant at <0.01 level
@ **MTR**=mean initial transpiration, **IC2**=initial cellular CO2 concentration, **RC2**=range of CO2 drop in chamber, **CHL**=leaf chlorophyll concentration, **MCM**=stomatal conductance, and **MPM**=rate of photosynthesis.

(Cladosporium caryigenum) has been reported by Gottwald and Wood (1985) to indicate the impact of a plant pathogen on the rate of MPM. Similarly, Basiouny and Biggs (1976) have reported a 43% decrease in the rate of peach leaf MPM due to zinc deficiency. The seasonal reduction in MPM from a higher rate in spring to the lowest in the fall which was observed in this investigation, may have resulted from the effect of leaf age in addition to the influences of such factors as reduced light intensity, photoperiod, and upper limit of ambient temperature (Inada 1976, Thomas 1955). In addition to MPM, area concns of CHL were decreased with the advance of growing season, but not as consistently as did the MPM rate although both of these were significantly correlated. However, few reports have also indicated that leaf aging caused no decrease in the CHL contents (Marini 1986) but shading effectively increased CHL in peach leaves (Kappel and Flore 1983). In fact, our data for microplot trees (Figs. 1-A, 2-A, 3-A) showed an increase in CHL concn as growing season advanced; however, this was not true in the other plantings which had older trees than in the microplots.

EXPERIMENT 1:

Over the period of seven months, the average rate of peach leaf MPM was 13.78 μmol CO_2 m^{-2} s^{-1} with a range between 20.72 μmol CO_2 m^{-2} s^{-1} in May to 7.89 μmol CO_2 m^{-2} s^{-1} in November. Type of RSTK significantly influenced rates of MTR, MCM and MPM (Figs. 5-C, 5-D, 5-E). The average values of CHL and IC2 concns and the rates of MTR, MCM, and MPM (Figs. 5-A, 5-B, 5-C, 5-D, 5-E) for 'Redhaven' trees on 'Lovell' RSTK were higher than those on 'Siberian C' RSTK. Ferree and Barden (1971) have reported such differences in MPM of apple trees which resulted from the RSTK effect. However, research of this nature in peach is lacking. The effect of phytohormones (PHMN) on MPM (Fig. 5-F) and also on other GEA related parameters was quite inconsistent as well as markedly weaker than the influence exerted by the RSTK type. Furthermore, these differences in the MPM rates and other GEA parameters caused by the influence of PHMN, were statistically inseparable. This could have been due to the fact that the PHMN effects are short-lived and do not persist longer than few days to several weeks. Since the plant growth-regulating substances occur naturally, exogenously applied PHMN may be degraded to less effective substances in the plant body. Such natural growth-inhibitory phytohormones as ABA and its close relatives have been reported to reduce MPM rate in the manner of antitranspirants mostly through their influence on stomatal regulation (Del Valle et al. 1985, Kriedemann et al. 1975, Zelitch 1962). However, this inhibitory effect of low ABA concn levels off with the passage of time and thus, higher ABA concns hold down the MPM rates longer than do the lower concns of this PHMN (Del Valle at al. 1985).

Neither RSTK nor PHMN showed consistent effects on all of the GEA parameters. There are many reports (Kriedemann et al. 1975, Ryc and Lewak 1982) that indicate synergistic influence of some PHMN on GEA parameters as well as on photosynthetic apparatus. In the present investigation, such effects due to exogenously applied PHMN were not observed (Figs. 3-A, 3-B, 3-C, 3-D, 3-E). Table 1 clearly shows that significant correlations existed between most parameters (MTR, IC2, MCM, MPM) and CHL. However, CHL values were not correlated with those of IC2 which showed a negative correlation with MPM rate. Since photosynthetic response for the leaves of C3 plant species usually levels off at IC2 values in excess of 250 - 270 μl L^{-1}, maintaining the concns of IC2 between 230 to 260 μl L^{-1} range maximizes their carboxylation potential while minimizing the MTR (Farquhar et al. 1980). A comparison of five 'Redhaven' trees on 'Siberian C' RSTK including a normal healthy and 4 other trees which succumbed to PTSL before or during winter/spring 1986 (Fig. 6-D), indicated that the single leaf MPM of the tree that was eventually collapsed before the end of 1985, was markedly reduced early in the season. Additionally, the response of these trees followed a similar general trend for leaf concns of CHL and the rates of MTR and MCM (Figures. 6-A, 6-B, 6-C). Both RSTK and PHMN responses showed a significant interaction for IC2, although their individual responses were not consistently significant during the period of this investigation.

EXPERIMENT 2:

This experiment was designed to study the PTSL development in 'Redhaven' peach trees grafted on two RSTKs which had variable cold hardiness and were growing in PTSL and non-PTSL soils under the same microclimatic conditions. Before monitoring of GEA data, these trees received two applications of 100 μg L^{-1} each of ABA and GA_3,and distilled H_2O was used as a check. Seasonal mean of monthly rate of net photosynthesis for this experiment was 12.84 μmol CO_2 m^{-2} s^{-1} with a wide range varying from 18.29 μmol CO_2 m^{-2} s^{-1} in May to 9.19 μmol CO_2 m^{-2} s^{-1} in October.

None of the 3 treatments (soil, RSTK, or PHMN) in this experiment, showed consistently significant influence on any of the GEA parameters. But RSTK had the greatest effect on most of the parameters, and this was followed by the influence of soil and then PHMN. Rates of MCM and MPM were significantly affected by RSTK type; however, this effect on MTR was less evident. As a matter of fact, trees on 'Nemaguard' were more efficient in assimilating CO_2 than those on 'Lovell' RSTK (Figs. 2-C, 2-D, 2-E). Since 'Nemaguard' when used as a RSTK, causes an increase in the scion vigor, it seems that a part of this invigorating effect of 'Nemaguard' may possibly be responsible for greater photosynthetic rate on this germplasm. A concluding evidence in this regard was presented by Chalmers et al. (1975) who indicated a strong coupling between MPM rate and the growth requirements of a peach tree. On the other hand, very little or no differences in MTR of leaves from the same peach cultivar on different RSTK may probably be apparent because the stomatal regulation plays a major role in the transpiration process. These results agree with the findings of a recent study on red- and green-leaf peach cultivars which showed no significant differences in MTR, MCM, and MPM (Marini 1986). There were no significant differences in CHL and IC2 due to these RSTK (Figs 2-A and 2-B). The

generally accepted fact that the induced closure of the stomates reduces MTR rate relatively more than the rate of MPM (Zelitch and Waggoner 1962), appeared to be evident in our peach tree study in the microplot experiment.

The effects of soil on GEA though not consistent from month to month or statistically significant, were quite interesting. There was very little difference in MTR (Fig. 1-B), while the variations in leaf CHL, IC2 and MCM (Figs. 1-A, 1-C, 1-D) were quite inconsistent. On the other hand, the MPM rate was almost invariably higher for trees on non-PTSL than for those on PTSL soil; however, the differences were not statistically significant (Fig. 1-E). As in the previous experiment, the response of PHMN was significant only for the IC2 concns in the leaf cells (Fig. 3-B). From these data, it appears that the exogenously applied ABA had prolonged effects on the stomatal regulation thereby keeping them incompletely open, and hence, resulting in the increased levels of CO_2 in the leaf cellular spaces. This has been suggested by several reports describing the close relationship of stomatal regulation with moisture stress which may have contributed to the build up of this (ABA) inhibitory PHMN (Bradford 1983, Kriedemann et al. 1975, Tan and Buttery 1982, 1983).

Comparison of MPM (Fig. 6-F) and MCM (Fig. 6-E) rates of leaves from 2 apparently healthy trees grafted onto 'Nemaguard' RSTK with those from an unhealthy and a PTSL-killed (in 1986) tree, showed clear differences in these parameters. It is also revealed from these figures that the apparently sick-looking tree which eventually died during the 1986 winter/spring, had significantly lower rates of MPM and MCM than healthy trees which maintained their normal health during 1985 season and grew vigorously like normal trees during the succeeding growth season of 1986. This decrease in the GEA associated with PTSL-related stress(es) may be similar to a decline in the MPM rate proportional to the colony size of the fungus responsible for pecan scab reported by Gottwald and Wood (1985). It is quite possible that other such factors like improper stomatal regulation and the **invisible** or **metabolic changes** resulting from certain stresses and unhealthy condition, may have contributed to some degree in the reduction of these GEA values. Significant coefficients of correlation between MPM and other GEA parameters for this study are presented in Table 1. The rate of MPM was highly correlated with the range of CO_2 (RC2) consumption (r=0.85) and MCM (r=0.48) but less so with MTR and CHL concns. Buttery and Buzzell (1977) reported that the area concns of leaf CHL and MPM rate have not always been closely related as being indicated by observations in this study.

EXPERIMENT 3:

Since 'Amaru' and 'Baim' peach cultivars incurred greater than 60% tree mortalities through the end of 1985 growing season and the tree survival of these two lines was further reduced during 1986 (Fig. 4-A) to less than 10% and 35% for 'Amaru' and 'Baim', respectively, these two cultivars were not included in the GEA observations reported in this paper. Furthermore, Figure 4-A showed that 'Wild peach' had 100% trees surviving by September 1986 in comparison to only 95% tree survival of 'Lovell' seedlings. This investigation where we compared gas exchanges performance of 'Wild peach' with that of 'Lovell' seedlings had an average photosynthesis rate of 13.94 μmol CO_2 m^{-2} s^{-1} which seasonally declined from a high of 19.02 μmol CO_2 m^{-2} s^{-1} in the month of May to a low of 8.25 μmol CO_2 m^{-2} s^{-1} in November. These data indicate that the type of germplasm significantly affected all GEA parameters except IC2. Similar variations among different stone fruit species (De Jong 1983), red and green leaf cultivars of peach (Marini 1986), and various rootstocks and some strains of apple (Ferree and Barden 1971) with regard to certain GEA have been documented in the literature. It was generally observed that the seedlings of 'Wild peach' showed a significantly better performance in GEA than did the seedlings of 'Lovell' peach (Figs. 4-D, 4-E, 4-F). In addition, the statistical differences in this study were more consistent for most of the parameters except IC2 concns (Fig. 4-C) than in the two experiments described above . On the other hand, leaves from 'Lovell' seedlings had higher concns of CHL than 'Wild peach' leaves (Fig. 4-B), but CHL was not statistically correlated with MPM (r=.141) as shown in Table 1. The effect of the germplasm types on CHL concns was very highly significant (p=<.0001) and stayed almost consistent over the entire 1985 season. The rate of photosynthesis was significantly correlated with all other GEA parameters except the chlorophyll concentrations.

CONCLUSIONS AND FUTURE DIRECTIONS

On the basis of one season's single leaf GEA data for seven consecutive months of vegetative growth period, gas exchange performances of peach trees were not consistent for the entire season. This mostly resulted from variable weather conditions, particularly high heat and lack of precipitation during late spring. Rootstock and cultivar types influenced photosynthesis and other GEA parameters more frequently than did the soil and phytohormone types. However, phytohormonal treatments showed significant influence on cellular CO_2 concns which were negatively correlated with the rates of MPM, MCM and MTR in many case. Patterns of several GEA parameters as influenced by tree health indicated that the reductions in these GEA responses were brought about by the PTSL-related stress of trees. These findings will be further investigated in the field through continued monitoring of gas echange responses of the same trees utilized in 1985 for at least 2 more growing seasons. Various kinds of stresses will be imposed on different types of peach germplasm under controlled conditions, and their influence on the physiological performance of trees will be investigated through gas exchange responses of these germplasm.

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BIOCHEMICAL CHANGES IN PEACH TREES ASSOCIATED WITH PEACH TREE SHORT LIFE

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Peach Tree Short Life (PTSL) is a complex interaction of factors which ultimately results in tree death of what appeared to be healthy peach trees the previous season. We endeavored to determine the biochemical and physiological responses of apparently "healthy" trees as they were subjected to stimuli associated with PTSL to determine the sequence of events leading to tree death in the spring of the year.

We compared "healthy" trees to those affected by PTSL in an orchard situation or trees exposed to various stimuli in the greenhouse. Some of the comparisons made were: fall versus spring pruning; rootstocks; presence or absence of the ring nematode (Criconemella xenoplax); high versus low soil pH; effects of temperature alternations as well as various combinations of the above factors. Physiological and biochemical measurements including determining the concentration and changes in prunasin, reducing sugars, ninhydrin positive materials, amino acid concentration and composition, phenols, leakage of electrolytes, evolution of cyanide, enzyme activity and protein profiles were made on the test material.

Interpretation of our data indicated that the rapid breakdown of prunasin due to changes in permeability with the concurrent evolution of cyanide was probably directly related to tree death in the PTSL syndrome. Presented here are the data on which our conclusions were derived.

Initially, consider the sequence of the degradation of prunasin in peach tissue. This cyanogenic glucoside is thought to be compartmentalized within the cell vacuole and kept isolated from β-glucosidase and mandelonitrile lyase, the enzymes which degrade the molecule (Conn 1980) (Fig. 1). Both the intermediate mandelonitrile and the end product cyanide are potent inhibitors of oxidation/reduction reactions in peach tissue.

An example of such is nitrate reductase inhibition by mandelonitrile and cyanide (Reilly et al. 1986) (Table 1).

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Prunus species in general and peach in particular have a potential for releasing cyanide from prunasin in far greater concentrations than necessary for the inhibition of physiological processes (Reilly et al. 1985) (Table 2) (Reilly et al. 1986) (Fig. 2). Prunasin contains 8.8% cyanide; therefore, 88 μg cyanide per mg of prunasin are released on the breakdown of the molecule. This amount of cyanide from 1-10 mg of prunasin depending on the tissue source, confined to less than 1 cm^2 of bark tissue would be highly toxic.

In greenhouse studies, infestation of ring nematode on 'Lovell'or 'Nemaguard' rootstocks significantly (P=0.01) increased prunasin content of both rootstocks whereas the content in stem tissue was decreased (Okie et al. 1984) (Table 3). In orchard studies, comparison of prunasin concentration of PTSL trees and adjacent healthy trees in the same orchard showed that the PTSL trees had a significant (P=0.01) decrease in prunasin content from trunk tissue and that the greatest decrease was from the south side of the tree (Reilly et al. 1986) (Table 4). The nematode populations under the PTSL trees were always significantly greater (P=0.01) than those under the adjacent healthy trees (Reilly et al. 1986) (Table 5).

To this point we could conclude that; a) Prunus species had a concentration of the cyanogenic glucoside prunasin in levels that when broken down, would release highly toxic amounts of cyanide.

b) Criconemella xenoplax was capable of altering the level of prunasin in both roots and stems.

c) PTSL affected trees had extremely low levels of prunasin in the trunk as opposed to healthy adjacent trees and that this low level was more evident on the south side of the PTSL tree.

d) Prunasin, was the most affected chemical component which we measured in PTSL trees.

However, from these studies we could not determine whether the breakdown of prunasin was the result of tissue damage or its cause. To determine the sequence of events leading to prunasin release and cyanide evolution, two studies were initiated. The first consisted of establishing a new orchard on a pre-existing short-life site. Treatments were lime/no lime x 'Lovell'/'Nemaguard' x fumigation/no fumigation x December pruned/March pruned.

In this study we found that prunasin levels in PTSL trees appeared to change just prior to or concurrently with the onset of damage to the tissue. Monthly samplings of root and stem tissue for prunasin, reducing sugars, ninhydrin positive materials, phenols, enzyme activities and protein profiles were conducted. Of the 16 treatments, only the fall pruned, non-fumigated 'Nemaguard' rootstocks showed decreased reducing sugars and prunasin but only on the last date sampled in both the limed and non-limed plots.

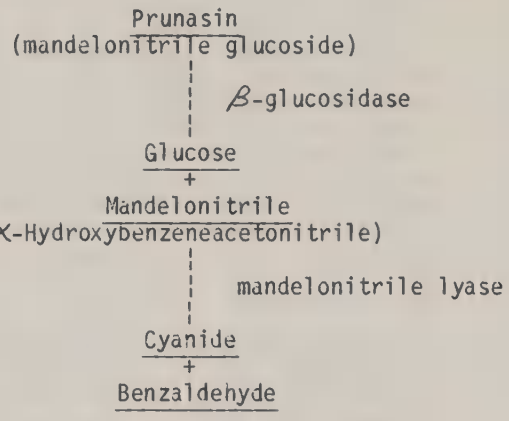

Figure 1. Degradation of prunasin by β-glucosidase and mandelonitrile lyase.

Table I. Effects of amygdalin and its breakdown products on nitrate reductase (NR) activity.

Sample[1/]	NR activity (nM NO_2/min/mg protein)	Percent Inhibition
NR assay untreated	1.55	--
Amygdalin	1.75	0
Prunasin	1.55	0
Mandelonitrile	0.17	89
Cyanide	0.00	100
Benzaldehyde	1.58	0
Acetone (10 ul)	1.67	0

1/ Ten micrograms of amygdalin or its breakdown products were added to the standard corn NR assay. Mandelonitrile and benzaldehyde were added in 10 ul of acetone.

Table II. Prunasin content of twig and trunk samples from 'Elberta' peach trees on 'Nemaguard' rootstocks.

Sample type	No. samples/tree	Prunasin (mg/g fresh weight ± SD)
	Twigs[1/]	
Elberta/Nemaguard (single trees)	3	9.04 ± 0.57
	3	7.41 ± 1.58
	3	8.16 ± 1.13
Elberta/Nemaguard (16 trees)	1	8.98 ± 2.03
	Trunk[2/]	
Elberta/Nemaguard (single trees)	3	2.98 ± 0.16
	3	2.06 ± 0.05
	3	2.02 ± 0.17
Elberta/Nemaguard (15 trees)	1	3.22 ± 0.74

1/ Pooled samples using single segments from each of 4 twigs per tree.
2/ Consisted of 1.3 cm diameter cylinders from the bark down to the xylem, which was discarded.

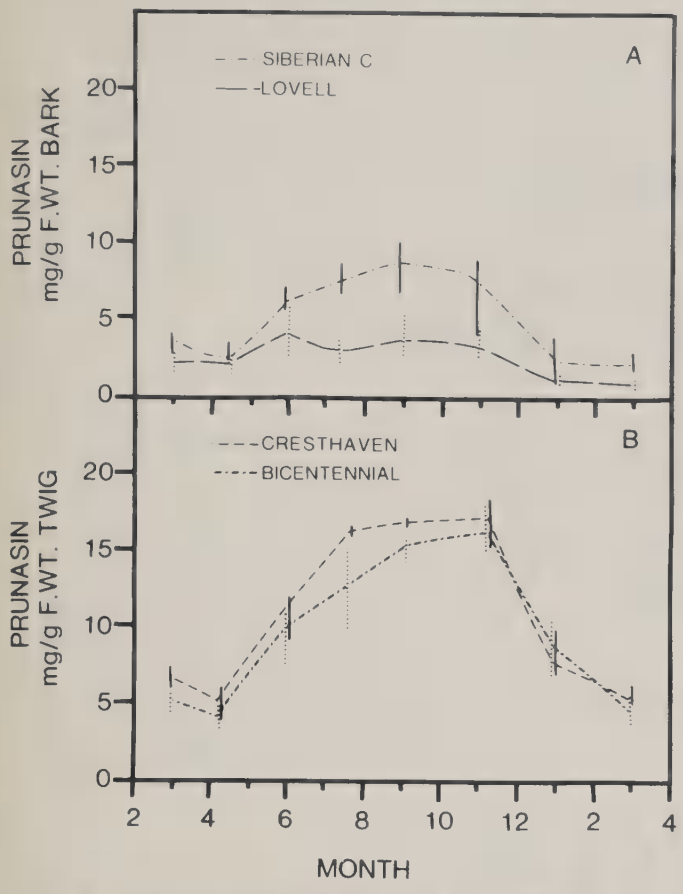

Fig. 2 Seasonal distribution of prunasin in the (A) root bark of 'Siberian C' and 'Lovell' rootstocks and (B) twigs of 'Cresthaven' and 'Bicentennial' varities.

Table III. The effect of 8 mo of exposure to Criconemella xenoplax (Cx) on the cyanogenic glucoside prunasin in peach seedlings and rooted herbaceous cuttings.

Rootstock	Cx	Prunasin(mg/g dry wt ± S.E.)[1/] Stem	Root
Herbaceous cuttings			
Lovell	-	6.7 ± 1.0	21.5 ± 2.7
Lovell	+	6.0 ± 1.0	26.2 ± 1.9
Nemaguard	-	5.2 + 0.5	16.8 + 4.0
Nemaguard	+	3.1 ± 0.3	23.8 ± 2.5
Seedlings			
Nemaguard	-	6.4 ± 1.0	15.9 ± 1.5
Nemaguard	+	4.2 ± 0.6	20.4 ± 1.4
Significant effects[2/]			
Cx		*	**
Rootstock		*	+

1/ Mean of six determinations per treatment.
2/ F-test significant at P=0.10 (+), 0.05 (*), or 0.01 (**).

Table IV. Mean prunasin content of peach trees affected by peach tree short life (PTSL), from apparently healthy trees in the same orchard, and from trees in non-PTSL orchards.

Orchard and type	Prunasin content (mg/cm^2 of bark)[1/] (±SD)			
	PTSL trees		Healthy trees	
	South	North	South	North
1 (PTSL)	0.09±0.20	0.95±0.31	1.02±0.50	1.40±0.48
2 (PTSL)	0.09±0.27	0.22±0.44	0.93±0.31	0.99±0.36
3 (PTSL)	0.00	0.00	1.28±0.27	1.33±0.56
4 (PTSL)	0.03±0.09	0.58±0.71	1.66±0.45	1.45±0.51
5 (PTSL)	0.00	0.44±0.66	1.04±0.36	1.64±0.46
6 (Healthy)	-	-	1.30±0.60	1.16±0.54
7 (Healthy)	-	-	1.82±0.54	1.66±0.34
8 (Healthy)	-	-	1.16±0.56	1.32±0.44
9 (Healthy)	-	-	1.46±0.72	0.92±0.14

1/ Samples of bark tissue were taken from the north and south sides of each tree 20-30 cm above ground level. Ten trees of each type were sampled in PTSL orchards and six trees in healthy orchards.

Table V. Number of Criconemella xenoplax under peach tree short life (PTSL) trees and healthy trees in PTSL or non-PTSL orchards in Georgia in April 1984.

Orchard and type	No. trees/ orchard	Percent PTSL	C. xenoplax/100 cm^3 of soil	
			PTSL trees[1/]	Healthy trees[1/]
1 (PTSL)	638	11.0	346	237
2 (PTSL)	3,248	28.0	356	139
3 (PTSL)	780	30.4	293	170
4 (PTSL)	921	27.5	158	134
5 (PTSL)	664	17.3	810	67
Mean		22.8	392**[2/]	149**
6 (Healthy)	496	0.6	-	127
7 (Healthy)	935	0.5	-	3
8 (Healthy)	780	1.5	-	362
9 (Healthy)	700	0.3	-	109
Mean		0.7	-	150

1/ Mean of 10 trees (PTSL-type orchard) or six trees (Healthy-type orchard).

2/** = Means of PTSL trees and healthy trees in PTSL orchards are significantly different (P=0.01).

In concurrent studies which were initiated to determine the effects of temperature fluctuations on trunk tissue, we reduced or increased this effect with insulation material. First the trees were pruned in December, then Armaflex pipe insulation was installed on the trunk to the scaffold branches, or trunks were wrapped with a clear plastic packing material. Comparison treatments were uninsulated, December-pruned or March-pruned trees.

Temperature profiles for the bark of the trees so treated were radically different both on sunny days and cloudy days (Fig. 3). The pipe insulation decreased temperature fluctuation to only a few degrees whereas the clear plastic wrap gave a wide range of temperatures over a 24-hr period.

The intensity of cambium browning has been a measure of degree of injury for peach trees (Yadava 1984). In this study the cambium of clear plastic-wrapped trees had the darkest cambium followed by the December-pruned, uninsulated trees, with the Armaflex-insulated trees showing the least browning (Table 6).

Another measure of tissue injury of dormant trees has been leakage of electrolytes as determined by changes in electrical conductivity. The plastic wrapped trees again showed the greatest changes both at room temperature (22 C) and 50 C (Table 7). This change in conductivity under experimental conditions indicated the plastic wrapped trees were in a less cold hardy condition and more susceptible to tissue damage or that damage had already occurred.

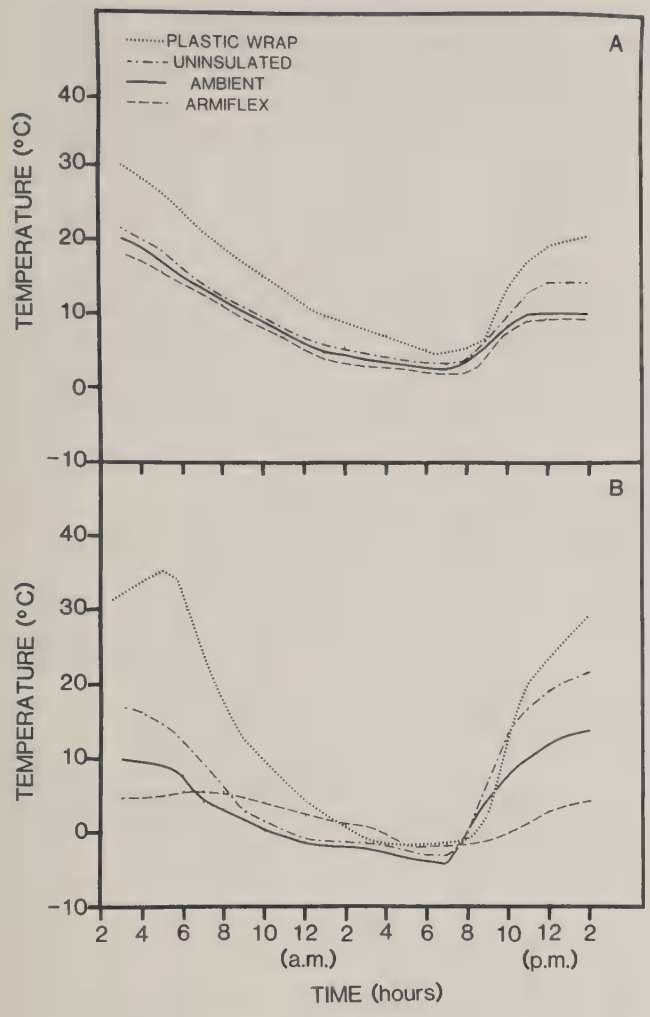

Figure 3. The temperature profile of peach tree bark under various insulation treatments.

The amount of prunasin available for rapid release of cyanide was also significantly greater (P=0.01) for the plastic-wrapped trees, even at the room temperature treatment (Table 8).

Apparently the trunk cells of the plastic-wrap treatment were less dormant and responded to fluctuations in air temperature by increased permeability changes; the loss of prunasin and increased conductivity probably also increased cambium browning in the orchard.

We therefore propose that damage to trunk tissue in PTSL occurs following 1.) alterations of permeability of membranes due to unfavorable stimuli such as pruning in December and/or nematode infestation-the effect being greater with both 2.) rapid and large temperature fluctuations which adversely affect the membranes mainly on the south to southwest side of the trunk where solar radiation is the greatest causing the predisposed cells to release prunasin and increase their permeability and 3.) the prunasin reacts with the appropriate enzymes to release cyanide which leads to more tissue damage and eventual death. The later 2 events occur in late winter - February or early March, at a time when the damage is not readily detectable.

Table VI. Cambial browning rating for March-pruned, December-pruned, December-pruned insulated and December-pruned plastic-wrapped trees, March 1986.

Treatment	Trunk Rating[1/]	
	South facing	North facing
March-pruned	1.87	0.87
December-pruned	3.50	3.00
December-pruned Insulated	0.25	0.50
December-pruned Plastic-wrapped	6.50	2.75

1/ Mean of 8 replicates, rating scale 0 = no browning to 9 = dark brown

Table VII. Effects of insulation on conductivity (March 1986).

Treatment	Percent Conductivity[1/]		
	+50 C	RT (22 C)	-25 C
March-pruned	38.09	43.54	72.34
December-pruned	39.97	47.47	76.89
December-pruned Insulated	39.07	45.03	76.79
December-pruned Plastic-wrapped	61.56	60.77	74.91

1/ Percent conductivity = μmho pre autoclave/μmho post autoclave

Table VIII. Insulation effects on rapid CN release.

Treatment	CN(μg)/plug[1/]		
	+50 C	22 C	-25 C
March-pruned	21.78	1.95	101.28
December-pruned	24.66	2.84	77.07
December-pruned Insulated	18.74	9.19	109.66
December-pruned Plastic-wrapped	39.91	34.22**	137.4

1/ After temperature treatment and incubation 1 ml removed, treated with β-glucosidase, and CN measured.

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COMPARATIVE STUDIES BETWEEN CRICONEMELLA XENOPLAX AND OTHER PLANT-PARASITIC NEMATODES AND THEIR EFFECT ON GROWTH AND PHYSIOLOGY OF PEACH.

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INTRODUCTION

The ring nematode, Criconemella xenoplax (Raski) Luc & Raski, has been demonstrated to make peach trees more susceptible to peach tree short life (PTSL) (Nyczepir et al. 1983). Three Criconemella species were detected in a recent survey of PTSL orchards throughout the major peach growing areas of Georgia and South Carolina. These were C. xenoplax, C. ornata (Raski) Luc & Raski and C. sphaerocephala (Taylor) Luc & Raski (Nyczepir et al. 1985), with the former two species occurring in 100% of the orchards sampled. Other genera present in greater than 50% of the samples, included Pratylenchus spp., Tylenchorhynchus spp. and Meloidogyne spp.

Parasitic feeding habit and hosts differ between phytoparasitic nematodes. Criconemella xenoplax, an ectoparasite, is pathogenic on peach (Lownsbery et al. 1973, Nyczepir et al.1983) whereas C. ornata (Co) has been associated with agronomic crops and grasses (Johnson 1970, Motsinger et al. 1976, Ratanaworabhan et al. 1970). 'Lovell' rootstock was reported to be a poor host to C. ornata (Johnson 1970) but information regarding Nemaguard susceptibility is unavailable.

In peach roots, prunasin is the primary cyanogenic glucoside that breaks down under adverse conditions to yield cyanide, one of two metabolites that are toxic to animals and plants (Mizutani et al. 1979). According to Kaethler et al. (1982), hydrogen cyanide acts as a feeding deterrent in peach leaves to the oblique-banded leafroller (Choristoneura rosaceana (Harris) insect. Such a phenomenon may explain why 'Lovell' is a poor host for Co. The root-lesion nematodes, Pratylenchus spp., are migratory endoparasites. Several different species were tested for parasitism on peach, but only P. vulnus Allen & Jensen and P. penetrans (Cobb) Filip. & Stek. reproduced on the cultivars tested under greenhouse conditions (Barker 1973).

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Pratylenchus vulnus was the only lesion nematode related to rapid peach feeder root deterioration and tree vigor under field conditions in Georgia (Fliegel 1969). Other lesion nematodes detected in this field study included P. brachyurus (Godfrey) Filip & Sch. and P. zeae Graham but they were not associated with root damage.

The stunt nematode, Tylenchorhynchus claytoni Steiner, is also an ectoparasite. This nematode is thought to be parasitic on peach (Barker et al. 1973, Nyczepir et al. 1983), but its pathogenic capability has not been demonstrated. The present study was designed to: (1) determine if C. ornata is parasitic and pathogenic on 'Nemaguard' peach; (2) compare the degradative pathway of prunasin and detoxification of cyanide in C. xenoplax and C. ornata and (3) determine if Pratylenchus sp. and/or Tylenchorhynchus sp. affect peach growth and physiology (e.g. prunasin- and ninhydrin-reactive compounds) as previously demonstrated with C. xenoplax (Okie et al. 1984).

Pathogenicity and Biochemical Comparisons Between Criconemella xenoplax and C. ornata

Criconemella xenoplax (Cx) significantly reduced root volume (P = 0.10), dry stem weight (P=0.01) and height (P=0.01) of Nemaguard peach after six months when compared to the check (Table 1). On the other hand, C. ornata (Co) did not affect peach growth. Co did reduce (P=0.01) root volume and dry top weight of common bermudagrass compared to Cx.

The population density of Cx per 100 cm^3 soil was greater than Co on peach after four and six months (Table 2). The population of Cx increased from the fourth to the sixth month sampling whereas the Co population density remained relatively stable and low. The inverse was true regarding both nematodes on common bermudagrass, except that Cx reproduced more on grass than Co did on peach after six months.

Both Cx and Co nematode extracts reacted with p-nitrophenyl-β-D-glucopyranoside to form p-nitrophenol (Table 3). This reaction indicates that both nematodes appear to have the enzyme, β-glucosidase. This reaction was further substantiated by the cyanide assay (Table 3) which detected CN released from prunasin. Both nematodes were capable of metabolizing prunasin, to release cyanide. Cyanide detoxification occurred only from extracts of Cx and not Co by the enzymatic formation of β-cyanoalanine and hydrogen disulfide (Table 4). This reaction was linear for Cx and not Co after 30 minutes and did not occur if the extract was boiled or the essential substrate cysteine omitted.

Based on the results of this experiment, we conclude that 'Nemaguard' peach is a good host for Cx but not Co, whereas the inverse is true regarding common bermudagrass. However, Cx reproduced more on common bermudagrass than Co on peach which indicates that Cx would maintain itself on this grass in the absence of peach under field conditions. One explanation as to

why peach is a poor host for Co and not Cx is that this species is not capable of metabolizing cyanide. This study also stresses the importance of weed control and nematicide recommendation. Common bermudagrass is a very common weed in peach orchards and runs rampant if not controlled. Nematicide recommendations on the other hand are based on total ring nematode counts per volume of soil. If common bermudagrass is not controlled and soil sampling is conducted in such orchards, erroneous nematode information may be obtained if Co is present in high number and Cx low. This would possibly result in an added expenditure to the grower resulting from unnecessary nematicide application.

Pathogenicity and Biochemical Comparisons Between Criconemella xenoplax, Pratylenchus sp., and Tylenchorhynchus sp.

Pratylenchus sp.(Le), Tylenchorhynchus sp.(St) and Cx were all parasitic on peach as measured by the final nematode population recovered from the soil and the reproductive rate (R) (Table 5). However, Cx had a numerically higher R-value than either Pratylenchus (Le) or Tylenchorhynchus (St) sp. on peach in both soil media. This suggests that peach is possibly a better host for Cx than for Le or St or that reproduction occurs at a faster rate for this nematode compared to the others. The number f Le recovered from the root systems of peach was very small compared to soil populations (Table 5).

Percent tree survival differed between soil media and nematode treatments after trees were trimmed back to a uniform height (28.0 cm). Death occurred in only the sand:vermiculite medium and in the presence of Cx. Three of the five trees died following pruning, but prior to being moved outdoors in September whereas the other two died in April shortly after being brought back into the greenhouse following pruning. We do not feel this is a random phenomenon since it has been previously demonstrated (Nyczepir et al., in press). We think that it is related to an interaction between pruning, population density of Cx, root production, and/or possibly available carbohydrate root reserves.

The effect these different nematodes had on plant growth and physiology is presented in Table 6. In soil, Cx reduced (P=0.01) dry stem weight but increased (P=0.01) prunasin concentration in stem tissue. Le or St did not affect stem weight or prunasin stem concentration compared to the check. In sand:vermiculite, where tree death occurred, Cx and Le significantly (P=0.01) reduced stem weight compared to St and the check. There were no differences between Cx and Le. Le reduced dry root weight (P=0.05) compared to St and Ck, but not Cx. It should be noted that even though dry root weight was not significantly affected between treatments in the soil medium, numerical trends were identical to those in sand:vermiculite. The only physiological parameters that resulted in treatment differences were prunasin stem concentration and ninhydrin-reactive compounds (NRC) in root tissue (Table 6). Parasitism by Cx resulted in decreased stem prunasin (P=0.05) concentration and NRC (P=0.01) concentrations in roots compared to the other nematodes and check. These results substantiate earlier studies with Cx (Okie 1984). Le on the other hand, increased (P=0.01) NRC compounds in root tissue compared to the check and Cx. Significant differences between height increase, stem diameter, concentration of reducing sugars and phenolic compounds in stem and root tissue and prunasin concentration in root tissue were not detected in either potting media in this study.

CONCLUSION

In conclusion, our nematode reproduction and growth data indicate that Cx, St, and Le are all parasitic on Nemaguard peach. Our plant growth data regarding Le and Cx confirms previous studies (Barker et al. 1973, Fliegel 1969, Nyczepir et al. 1983), but this is a first report demonstrating parasitism of St on peach under controlled greenhouse conditions. Earlier studies which implicated St as a parasite of peach were results of statewide surveys (Barker (1973, Nyczepir et al. 1985) or microplot studies, (Nyczepir et al. 1983) where other vegetative hosts may have grown periodically to support St populations. The St nematode also did not affect any of the physiological parameters measured in this study; whereas Cx and Le did. This may have been the result of feeding site. Both Cx and St are characterized as ectoparasitic nematodes. The St nematode is primarily considered to be an epidermal feeder, whereas Cx feeds on both epidermal and cortical cells. The Le nematode is a migratory endoparasite and like Cx it primarily feeds on and destroys cortical root cells. Even though Le and Cx both reduced dry stem weight in sand:vermiculite-grown plants something additional occurred in the Cx inoculated plants causing them to die. Whether or not it was prunasin stem concentration or a reduction in ninhydrin-reactive compounds in root tissue is difficult to say based on the results of this study.

Lastly, is the question of the sand:vermiculite medium and its contributing role to tree death. If the medium itself was a direct or indirect cause of tree death, then one would expect other nematode-inoculated and check treatments in the same media to die; they did not. Nutritional status of the surviving plants was also in the acceptable range for peach (see Sharpe et al., in proceedings), however, we do not know if macro and micro nutrients may have been altered in those trees that eventually died prior to death. Possibly a time-lapse study would give us a better understanding and answers to the questions that have arisen from this test, assuming these compounds (e.g. prunasin) are involved with peach tree death.

Table I. Effect of Criconemella xenoplax (Cx) and C. ornata (Co) on Growth Parameters of Nemaguard Peach and Common Bermudagrass After 6 Months in the Greenhouse.

Trt	Root Volume (ml)	Dry Root wt. (g)	Dry Stem wt. (g)	Height Increase (cm)
1. CK-P[1/]	1.85	4.39	5.28	69.8
2. Cx-P	1.43	3.46	4.09	57.3
3. Co-P	2.13	5.13	5.54	70.7
4. Cx-G	2.68	3.32	8.61	-
5. Co-G	1.55	2.47	6.89	-
Contrasts				
CK-P vs Cx-P	+	NS	**	**
CK-P vs Co-P	NS	NS	NS	NS
Cx-G vs Co-G	**	NS	**	-

1/ P = Peach; G = Grass
2/ ** = P = 0.01; + = P = 0.10; NS = No significant differences

Table II. Population Density of Criconemella xenoplax (Cx) and C. ornata (Co) on Nemaguard Peach and Common Bermudagrass After 4 and 6 Months in the Greenhouse.

	Number Nematodes/100 cm^3 Soil			
	4 Month		6 Month	
Trt	Cx	Co	Cx	Co
CK-Peach	0	0	0*	0
Cx-Peach	7,771	0	29,532	0
Co-Peach	0	7	0**	7
Cx-Grass	70	0	564	0
Co-Grass	0	10,588	0	72,538

* One pot contaiminated with Cx
** Two pots contaminated with Cx

Table III. β-glucosidase Activity of Extracts From Criconemella xenoplax (Cx) and C. ornata (Co).

Treatment	Substrate		Activity	
	PNPG*	Prunasin	p-NP** (ug)	CN (ppm)
Cx	+		28.24	-
		+	-	0.50
Cx-boiled	+		0.00	-
		+	-	0.00
Co	+		27.35	-
		+	-	0.43
Co-boiled	+		0.00	-
		+	-	0.00
β-glucosidase	+		>120.00	-
		+	-	>10.00
β-glucosidase + boiled	+		0.00	-
		+	-	0.00
PNPG (check)	+		0.00	-
Prunasin (check)		+	-	0.00

* PNPG = p-nitrophenyl-β-D-glucopyranoside
** p-NP = p-nitrophenyl
CN = cyanide

Table IV. Reaction Time of β-Cyanoalanine Synthase in the Presence of Criconemella xenoplax (Cx) and C. ornata (Co) enzyme extracts.*

Time (min)	KCN	Cysteine	H_2S detected (µg) Cx	Co
0	+	+	0.00	0.00
5	+	+	0.47	0.00
15	+	+	1.42	0.00
30	+	+	2.61	0.24
30	+	-	0.00	0.00
30 (minus extract)	+	+	0.00	0.00
30 (extract + boiled)	+	+	0.00	0.00

*HCN + Cysteine $\xrightarrow[\text{synthase}]{\beta\text{-cyanoalanine}}$ β-cyanoalanine + H_2S

Table V. Population Density and Reproduction of Criconemella xenoplax (Cx), Pratylenchus Sp. (Le), and Tylenchorhynchus Sp. (St) and Their Relationship to Survival of Nemaguard Peach After 14 Months in Two Different Soil Media.

Trt	Soil[1/] Nemas/100cm³ Soil	R[2/]	% Tree Survival	Sand:Vermiculite (50:50)[1/] Nemas/100cm3 Soil	R	% Tree Survival
Cx	18,479	136.9	100	14,462	105.6	50
Le	2,061(5)[3/]	15.3	100	1,123(27)[3/]	8.3	100
St	5,091	37.7	100	863	6.4	100
Check	0	0	100	0	0	100

1/ Data are means of ten reps for soil medium and seven reps for sand:vermiculite medium.

2/ R = final nema. population divided by initial population (Pi); Pi = 5000 nemas/pot.

3/ Mean number of Pratylenchus sp. recovered/dry root system; in parenthesis.

Table VI. Influence of Criconemella xenoplax (Cx), Pratylenchus sp. (Le) and Tylenchorhynchus sp. (St) on Growth and Physiology of Nemaguard Peach After 14 Months in Two Different Soil Media.

Trt	Soil: Dry Stem wt.(g)	Dry Root wt.(g)	Stem Prunasin(mg)	Root Ninhydrin(mM)	Sand:Vermiculite(50:50): Dry Stem wt.(g)	Dry Root wt.(g)	Stem Prunasin(mg)	Root Ninhydrin(mM)
Cx	13.0a**	15.5a	8.3a**	3.9a	19.8a**	13.9ab*	3.1a*	1.4c**
Le	20.2b	14.6a	5.4b	5.1a	19.3a	11.0b	4.3b	5.3a
ST	27.0b	20.3a	5.9b	5.3a	22.2b	15.2a	4.4b	4.3ab
Check	23.8b	21.5a	5.2b	5.1a	23.8b	16.0a	5.2b	3.6b

Means with columns followed by the same letter do not differ significantly at P = 0.01 (**) or P = 0.05 (*) according to Duncan's New Multiple Range Test.

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EFFECT OF NEMATODES ON NUTRIENT UPTAKE IN LOVELL AND NEMAGUARD PEACH SEEDLINGS.

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Injury to plants by nematodes is usually evaluated by measuring changes in the host plant, generally reductions in growth or yield. The limited work that has been done on the relationship between nematodes and host nutrition indicates that nematodes have a profound influence on the uptake and accumulation of various nutrients. However, these influences vary according to host and nematode species involved. Rootknot nematodes (Meloidogyne spp.) have been found to decrease P concentrations in lima beans (Parris et al. 1943) but increase P as well as N and K in tomato roots (Maung et al. 1959). Potassium concentrations were increased in boxwood leaves due to Pratylenchus spp. infestation (Tarjan 1950) but P. vulnus decreased K in walnuts (Lownsbery 1956) and roses (Sher 1957). The effect of nematodes on nutrient concentrations can also vary with the plant part sampled. Jenkins and Malek (Jenkins et al. 1966) studied the effects of four different nematode genera on vetch and found that N was the most seriously affected root nutrient and K the most seriously affected shoot nutrient. Tarjan (1950) reported similar results in boxwood, with increased N levels in the roots and decrease K levels in the shoots due to nematode infestation.

Information on changes in peach nutrition due to nematode feeding is limited. Chitwood et al. (1952) found increased K and Ca and decreased Mg and Fe in peach leaves due to Meloidogyne spp. The magnitude of the change in nutrient concentration varied with cultivar as well as nematode species and the degree of infestation. Since there is little information available concerning the relationship between nematodes and peach nutrition, two greenhouse studies were conducted to determine the effect of four nematode genera on the nutrient status of peach seedlings.

MATERIALS AND METHODS

Experiment 1:

Uninoculated Nemaguard seedlings were compared with seedlings inoculated with ring [Criconemella xenoplax (Raski) Luc & Raski], stunt (Tylenchorhynchus sp.) or lesion (Pratylenchus sp.) nematodes. These genera were selected for use because they are found in peach orchards throughout the Southeast (Nyczepir et al. 1985) and because they differ in feeding habit from one another. Ring and stunt nematodes are ectoparasites and lesion nematode is a migratory endoparasite. Seven replications were in soil and ten replications in a 50:50 sand:vermiculite (S/V) mix. Nematodes were added at planting in May 1985. After 90 days, seedlings were cut back to 28 cm in height. In September, plants were moved to a lathhouse for accumulation of chilling hours. Seedlings were moved back to the greenhouse in April, 1986. The experiment was terminated in June (14 months).

Experiment 2:

Treatments were a factorial arrangement of two cultivars, (Lovell and Nemaguard) two levels of soil pH (5.5 and 6.3), and three nematode treatments. Nematode treatments were uninoculated checks, ring, or rootknot [Meloidogyne incognita (Race 3)] inoculated at 5000 nematodes/pot. Soil pH was adjusted with Ca $(OH)_2$. The experiment was terminated after 180 days.

RESULTS AND DISCUSSION

Experiment 1:

The influence of nematodes on plant growth and nutrient concentrations in Exp. 1 followed the same general trends in both soil and S/V; thus the data were combined for this presentation. Dry weight for leaves, roots and stems are presented in Figure 1. Ring nematode decreased leaf and stem weight by 33% and root weight by 19% compared to the uninoculated checks. Lesion nematode also decreased dry weight with the greatest decrease in root tissue. Stunt nematode did not significantly affect weight of leaves, roots, or stems.

Nutrient concentration data for Exp. 1 are presented on a total plant basis since the relative trends in leaves, roots and stems due to nematode inoculation were similar. In general, the macronutrient concentrations were reduced by ring and lesion and unaffected by stunt (Fig. 2). Concentrations of N, P, and K were reduced 12, 13, and 19% respectively by ring nematode compared with uninoculated check. Lesion nematode also decreased P, Ca and Mg concentrations. Stunt nematode decreased P but increased Mg concentrations over the levels found in the checks. Decreases in nutrient concentrations due to lesion and ring nematodes were relatively small, but decreases in total nutrient content were much more substantial. The relationship between percent concentration and total nutrient content is represented by plant N (Fig 2.). Lesion did not significantly affect N concentrations but did decrease total N content. Ring decreased N concentration by 14% but decreased total N content by 38%.

The effect of ring nematode on minor element concentrations was opposite their effect on macronutrient concentrations. That is ring

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nematode tended to decrease macronutrients but increase micronutrients. This effect was most pronounced for Al and Mn levels, increasing the concentrations of these elements 39 and 42%, respectively, as compared to the checks (Fig. 3). In contrast to ring, both lesion and stunt nematodes significantly decreased Al and Mn concentrations below the levels found in the checks (Fig. 3). Ring nematode also increased Fe, Cu, and Zn concentrations,but the 22% increase in Zn levels with ring was not significantly greater than Zn levels in the checks (Fig. 3). Both lesion and stunt decreased Fe, Cu, and Zn levels with lesion causing the largest decreases. The relationship between micronutrient concentrations and total micronutrient content is shown with plant Mn. Stunt and lesion nematodes decreased total Mn content but ring increased both total micronutrient content as well as concentrations (Fig. 3).

Stunt nematode did not affect plant dry weight but decreased minor element concentrations as compared with the checks. This would indicate that stunt is parasitic on peach, although the effects in this study were relatively minor. Lower elemental concentrations with lesion nematode may be related to reduced root mass and absorptive root area caused by the migratory endoparasitic feeding habit of this nematode. The effects of ring nematode on elemental concentrations are more difficult to explain. Ring nematode decreased root weight which should decrease elemental uptake. One possible explanation for increased minor elemental concentrations with ring nematode is soil pH. As soil pH decreases the availability of Al, Mn, Fe, Cu, and Zn increases, and ring nematode significantly decreased media pH in both the soil and S/V mix (Table 1). In the soil, pH decreased from 5.8 in the check to 5.3 in the ring inoculated pots; and in S/V, pH decreased from 7.4 in the checks to 6.9 in the ring nematode treatment. Neither lesion nor stunt nematode significantly influenced pH in either media.

Experiment 2:

There were differences in plant dry weights and nutrient concentrations due to high and low pH treatments and cultivars,but the data for these treatments will not be presented unless there were significant interactions between these treatments and nematodes. Dry weights of leaves, roots, and stems all followed the same general pattern in relation to the nematode treatments; thus only total plant dry weight are presented. There was a significant interaction among soil, cultivar, and nematodes for total plant dry weight (Table 2). With Lovell seedlings, the effect of both ring and rootknot was much greater at low soil pH than at the high pH. Dry weight of plants inoculated with ring and rootknot at low pH were significantly less than both uninoculated checks at low pH and inoculated plants at high pH. Soil pH did not have as great an effect with Nemaguard seedlings (Table 2). Ring nematode decreased plant weight below that of the checks at high pH but not at low pH. Rootknot nematode did not affect plant weight of Nemaguard seedlings.

The effect of soil pH and nematodes on N levels in Lovell seedlings was opposite their effect on plant weight (Table 3). The highest N levels in Lovell seedlings were with rootknot, followed by ring nematode at low pH. The increase in percent N with these treatments may be due to a concentration of N associated with reduced growth because ring nematode did decrease percent N at high pH when there was no difference in plant weight. With Nemaguard seedlings, ring nematode reduced N levels at both high and low soil pH as compared to the checks, although the decrease at high pH was not significant (Table 3).

Only total plant concentrations of K, Ca, and Mg are presented in Figure 4, since the relationships between nematodes and nutrient concentrations were similar for leaves, roots, and stems. Rootknot nematode increased K, decreased Ca, and had no effect on Mg concentrations as compared to the check. Ring nematode increased K concentrations but had no effect on Ca or Mg levels.

The influence of both nematodes on P accumulation varied with plant tissue (Fig. 5). Phosphorus concentration in the stem tissue was about 0.09% and was not affected by either nematode. Both ring and rootknot nematodes decreased P concentrations in the roots, but concentrations in the leaves were increased by rootknot.

Minor element concentrations also varied with nematodes and plant tissue. In the roots, Cu concentrations were decreased by rootknot (Fig. 5) and Fe by ring nematode (Fig. 6). In the stems, Al, Fe and Cu were decreased by ring and rootknot; Zn was decreased by rootknot alone; and Mn levels were increased by both nematodes (Figs. 5 & 6). In the leaves, nematode infestation tended to increase minor element concentrations. The increases in leaf Mn and Fe concentrations were due to increases in the low pH treatments. Ring nematode increased leaf Mn and Fe levels,and rootknot nematode increased leaf Mn levels above those found in the low pH check and all the high pH treatments (Table 4). The effect of ring nematode on soil pH was also greater in the low pH treatments (Table 1). In the low pH treatments soil pH decreased from 5.42 with the checks to 5.03 with ring nematode. In the high pH treatments ring nematode decreased soil pH,but the decrease was not significant at the 5% level.

The trends in Exp. 2 were not as clear as in Exp. 1. The effect of nematode infestation on nutrient concentrations in Exp. 2 varied with pH treatment and plant tissue, but some trends were apparent. The effect of nematodes was greater at low pH than at high pH, minor element concentrations in the leaves tended to increase with both nematodes, and ring nematode decreased soil pH. Differences between Exp. 1 and Exp. 2 may be due to the age of the plants. In Exp. 1, plants were 14 months old while in Exp. 2 plants were harvested after 6 months. Six months may

have been too short a time span for the effects of nematodes on nutrient uptake to fully manifest themselves. Or the severe cutting back in Exp. 1 may have enhanced the nematode effects. In any event, the data indicate that nematodes do influence the nutrient balance in peach trees, thus potentially influencing physiological processes.

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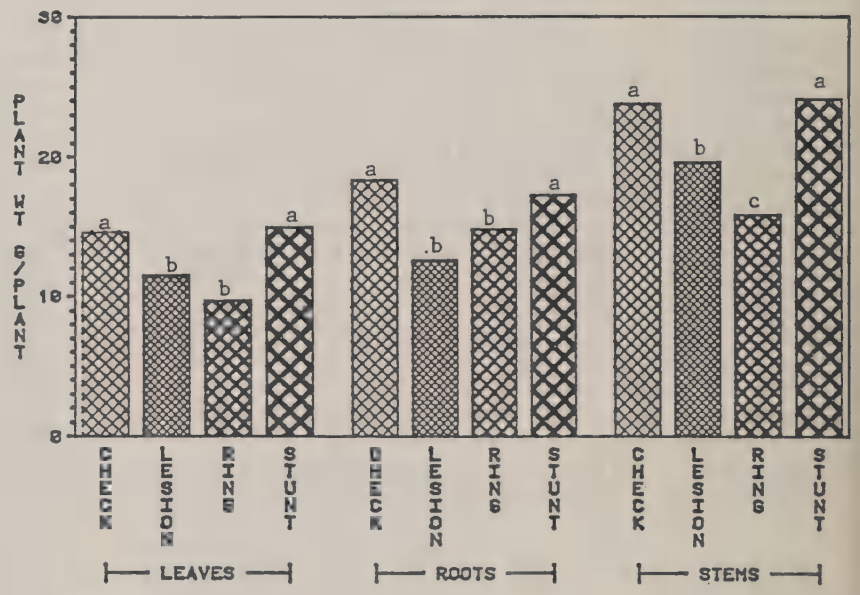

Fig. 1. Effect of nematodes on the dry wt. of leaves, roots and stems in Exp. 1. Means separation by Duncan's multiple range test, 5% level.

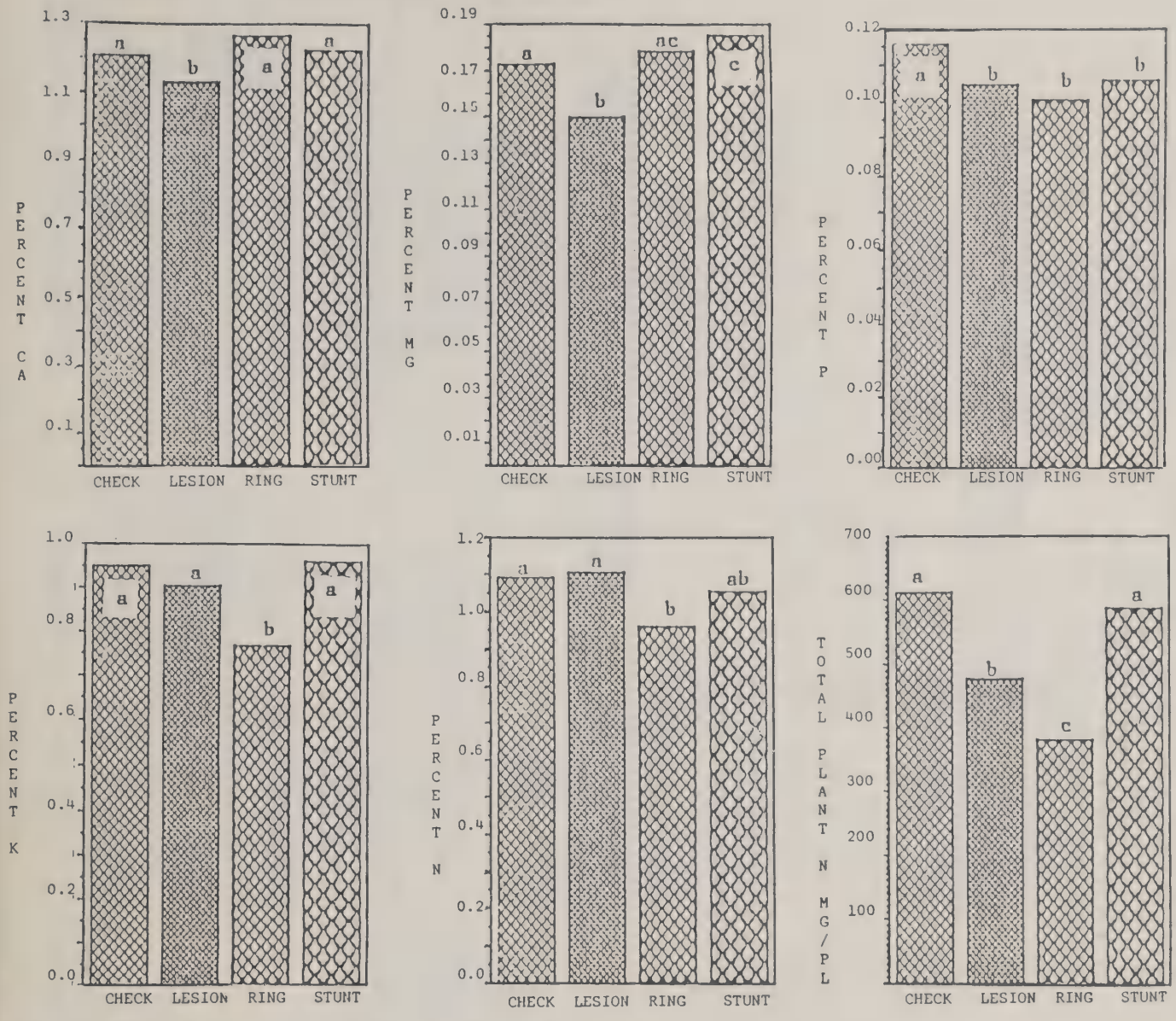

Fig. 2. Effect of nematodes on Ca, Mg, P, K, and N concentrations and total plant N content in Exp. 1. Means separation by Duncan's multiple range test, 5% level.

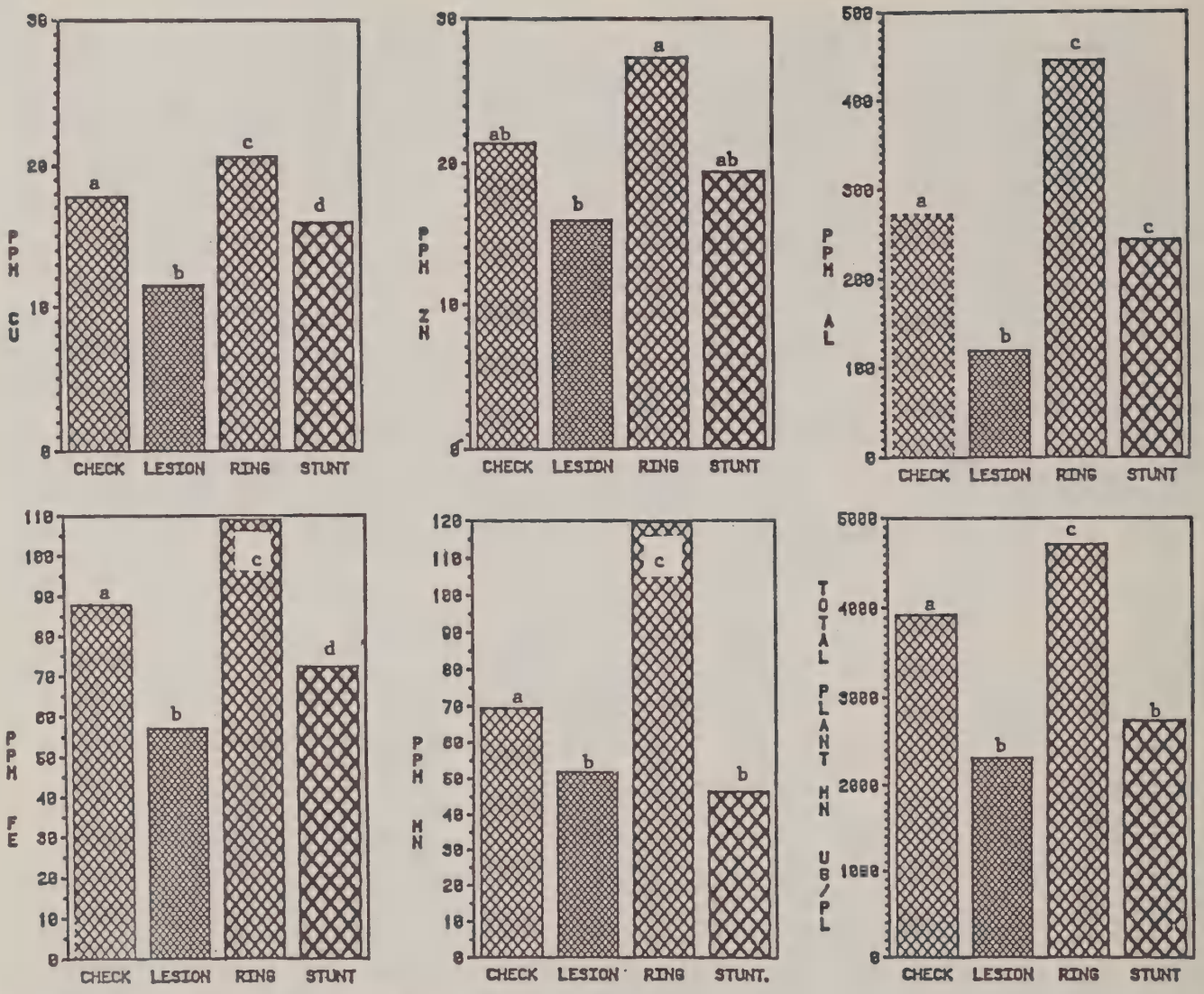

Fig. 3. Effect of nematodes on Cu, Zn, Al, Fe, and Mn concentrations and total plant Mn content in Exp. 1. Means separation by Duncan's multiple range test, 5% level.

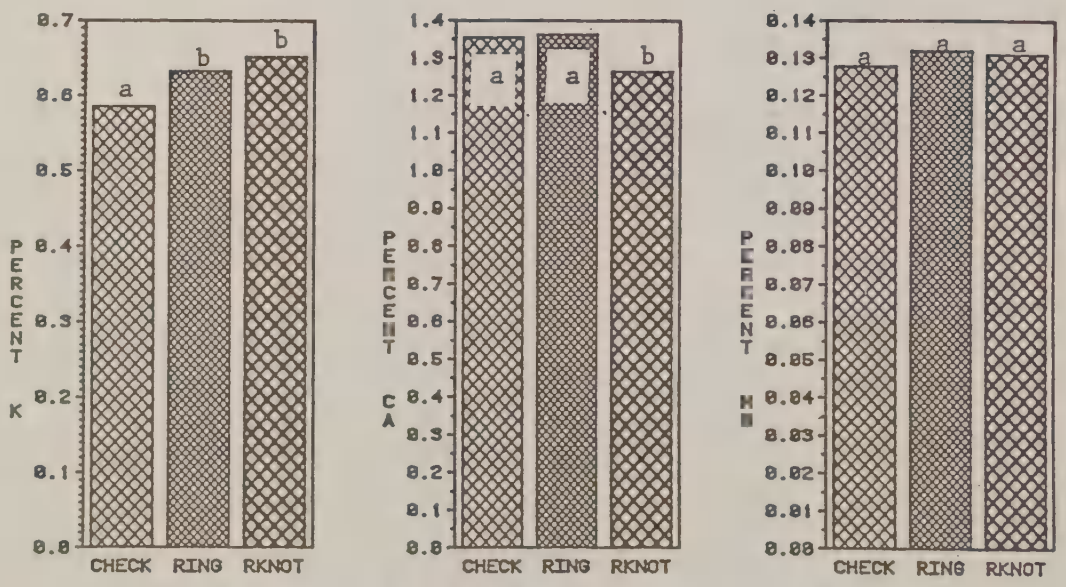

Fig. 4. Effect of nematodes on K, Ca, and Mg concentrations in Exp. 2. Means separation by Duncan's multiple range test. 5% level.

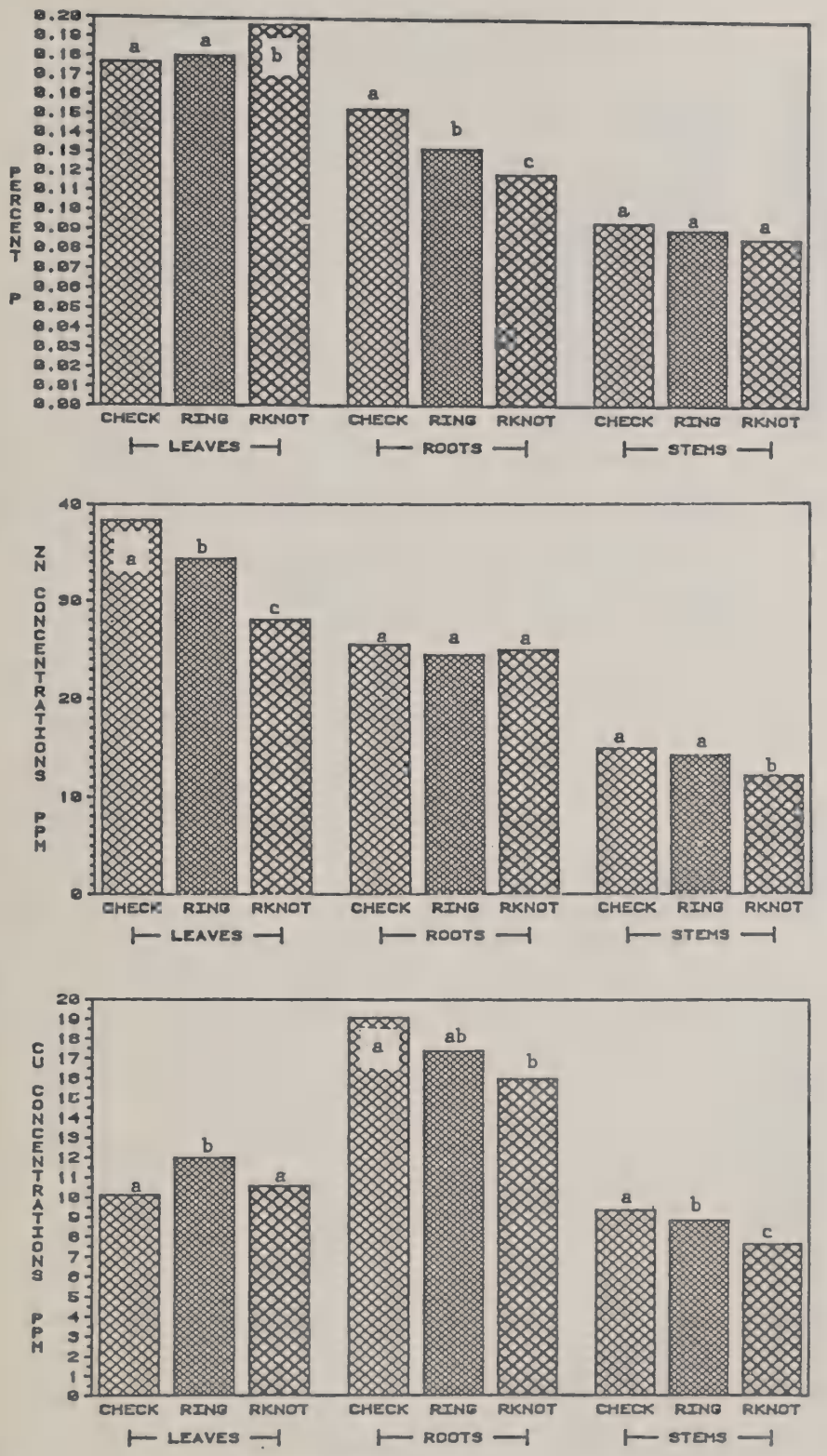

Fig. 5. Effect of nematodes on P, Zn, and Cu concentrations in peach tissue in Exp. 2. Means separation by Duncan's multiple range test, 5% level.

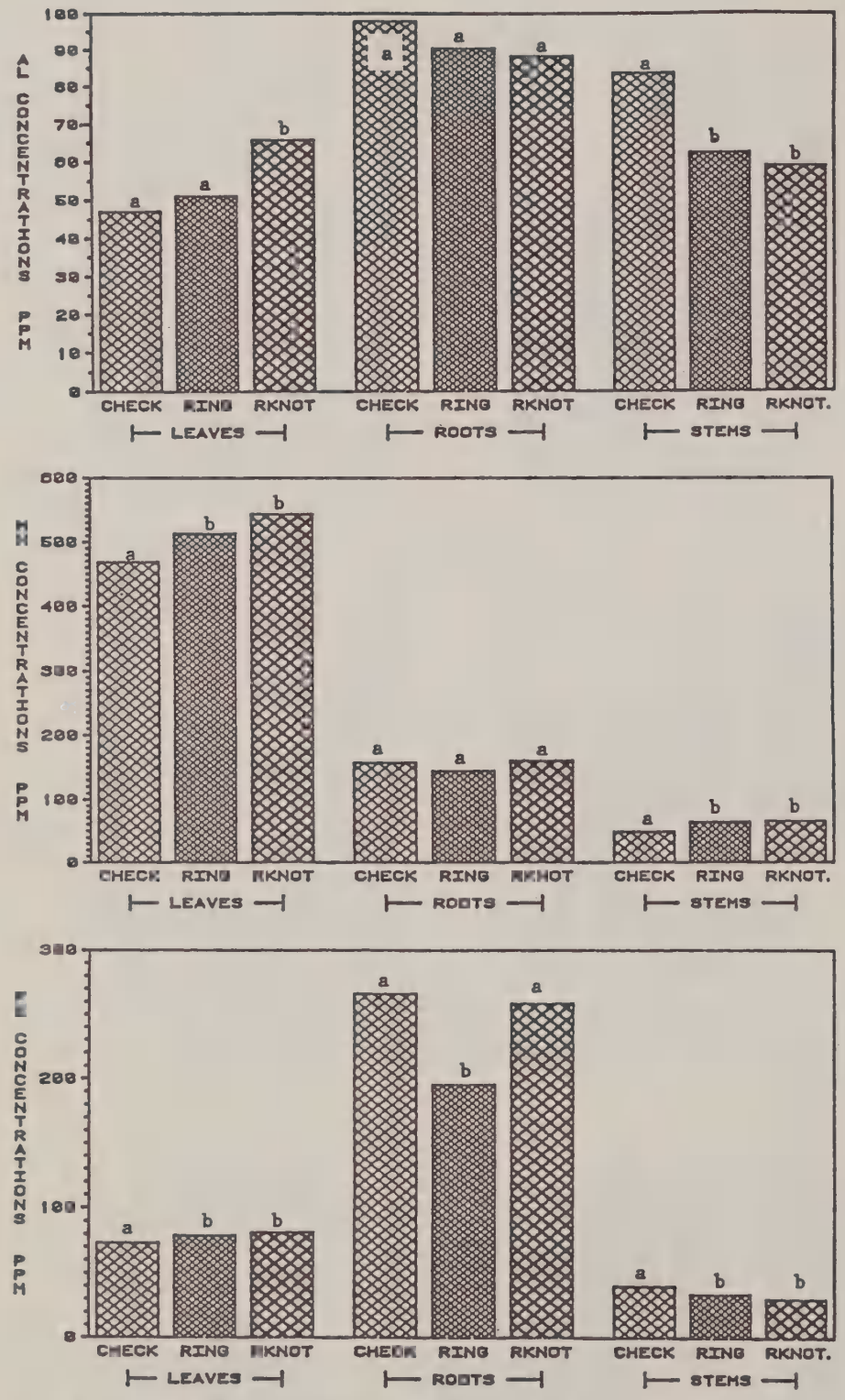

Fig. 6. Effect of nematodes on Al, Mn, and Fe concentrations in peach tissue in Exp. 2. Means separation by Duncan's multiple range test, 5% level.

Table I. Effect of nematodes on media pH.

NEMATODES	Soil[1/]	S/V[1/]	pH Treatment[2/] Low	High
Check	5.81 A[3/]	7.36 A	5.42 A	5.74 A
Stunt	5.89 A	7.03 AB	-	-
Lesion	5.96 A	7.29 A	-	-
Ring	5.28 B	6.95 B	5.03 B	5.40 A
Rootknot	-	-	5.59 A	5.69 A

1/ Growth media in Exp. 1.
2/ Soil pH treatments in Exp. 2.
3/ Mean separation within columns by Duncan's multiple range test, 5%.

Table II. Interactions of soil pH, cultivar, and nematodes on plant dry weight (g/plant).

	Lovell		Nemaguard	
Nematode	Low pH	High pH	Low pH	High pH
Check	43.55 Aa[1/]	40.13 Aa	46.74 Ab	52.42 Ab
Ring	27.49 Ba	39.26 Ab	44.11 Ab	40.87 Bb
Rootknot	21.98 Ba	32.83 Ab	45.25 Ac	48.28 Ac

1/ Mean separation by Duncan's multiple range test, 5% level. Upper case letters refer to mean separation within columns; lower case letters refer to mean separation within rows.

Table III. Interactions of soil pH, cultivar and nematodes on percent N in peach seedlings.

	Lovell		Nemaguard	
Nematode	Low pH	High pH	Low pH	High pH
Check	1.66 Aa[1/]	1.80 Aa	1.70 Aa	1.63 Aa
Ring	1.91 Ba	1.63 Bb	1.48 Bb	1.49 ABb
Rootknot	2.18 Ca	1.84 Ab	1.53 ABc	1.43 Bc

1/ Mean separation by Duncan's multiple range test, 5% level. Upper case letters refer to mean separation within columns; lower case letters refer to mean separation with rows.

Table IV. Relationship between soil pH and nematodes on leaf Mn and Fe concentrations.

	Leaf Mn (ppm)			Leaf Fe (ppm)		
pH	Check	Ring	Rootknot	Check	Ring	Rootknot
Low	559 B[1/]	679 A	708 A	74.8 b[z]	84.4 a	81.5 ab
High	392 C	347 C	388 C	71.9 b	73.6 b	82.4 ab

1/ Mean separation by Duncan's multiple range test, 5% level. Upper case letters refer to mean separation for Mn concentrations; lower case letters refer to mean separation for Fe concentrations.

CYTOSPORA CANKER

Paul F. Bertrand [1/]

INTRODUCTION

Cytospora canker, also known as perennial canker, Valsa canker or peach canker, is one of the most common stone fruit diseases in North America. The specific details that regulate the occurrence of Cytospora canker, i.e., winter injury, drought stress, etc. may vary within a region and certainly vary from one region to another. The role of Cytospora canker in stone fruit decline definitely varies from one region of North America to another. The concept of stone fruit decline itself varies among different regions in North America. However, granting these regional variations in predisposing factors, a search of literature and observations in various stone fruit regions indicates Cytospora canker as a disease varies little from one region to the next and could be thought of as being the same disease throughout the stone fruit culture of North America.

Cytospora canker of stone fruit is caused by two pathogenic species; *Leucostoma persoonii* (Nits.) v. Hohn. (=*Valsa leucostoma* Fr.), (anamorph *Cytospora leucostoma* Sacc.) and *Leucostoma cincta* (Fr.) v. Hohn. (=*Valsa cincta* Fr.), (anamorph *Cytospora cincta* Sacc.) (Kern 1955). The morphological differences between these two species as they occur in nature are often small, indistinct or overlap greatly (Kern 1955). Because of this, the two species generally have been separated on the basis of cultural characters defined by Willison (1936) or some modification of them. The *Cytospora* stages are seen far more frequently on diseased orchard trees than the corresponding *Leucostoma* stages. Observations during a study of Cytospora canker in California indicated that during the first two years after infection only the *Cytospora* stage was present in diseased tissues. Development of pycnidia is usually evident within six months after a canker is visible. The *Leucostoma* stage was seen only on completely dead branches two or more years after infection (Bertrand and English 1976b). *Leucostoma persoonii* was never encountered in an orchard where dead wood removal was a regular part of the pruning program. *Leucostoma persoonii* and its anamorph *Cytospora leucostoma* can be actively producing spores in the same canker (Bertrand and English 1976b) but the duration of activity of either stage in dead wood is not known.

Release and spread of conidia and ascospores was studied in California (Bertrand and English 1976b). Conidia and ascospores were released in response to rainfall any time of the year rainfall occurred. Conidia were carried in wind blown rain at least 76.8 meters from their source. Both conidia and ascospores were caught in rainwater running off sporulating cankers, suggesting localized splash dispersal mechanisms. Ascospores but not conidia were observed to be forcefully discharged into the air following a rain shower. These studies demonstrated effective air and splash dispersal mechanisms for *L. persoonii*. Spread of *L. persoonii* has also been shown to occur by infested pruning tools (Chiarappa 1960) and possibly by the shot hole borer (*Scolytus rugulosus* Ratz.) (Chiarappa 1960, DeVay 1974). Observations strongly suggest these latter means of spread are very minor as compared to spread in air, wind blown rain, and local washing and splashing. Studies of spore release and dispersal have not been conducted in the Eastern peach growing regions. However, other than the obvious seasonal rainfall differences between the central valley of California and the East coast the basic physical mechanisms should be the same.

The distribution of *Leucostoma persoonii* and *L. cincta* in the stone fruit growing regions of the United States is fairly well known (Alfieri *et al.* 1973, Bertrand and English 1976a, Endert 1985, Helton and Konicek 1961, Hildebrand 1947, Jones and Leupschen 1971, Rosenberger 1982, Wensley 1964, Willison 1936, Wysong and Dickens 1962). There is a well documented variation in virulence among isolates recovered from Cytospora cankers (Bertrand and English 1976a, Gairola and Powell 1970, Helton and Konicek 1961, Wysong and Dickens 1962). There is an equally well documented variation in resistance of various peach cultivars to *Cytospora* isolates (Gairola and Powell 1970, Luepschen 1981, Palmiter and Hickey 1970, Scorza and Pusey 1984, Wensley 1970, Wysong and Dickens 1962). Even though there are differences among peach cultivars in resistance to Cytospora canker there does not appear to be any strong evidence for a level of resistance that would equate to a means of control. Comparative studies of relative cultivar resistance to Cytospora canker is lacking for other *Prunus* sp.

The status of *L. persoonii* and *L. cincta* as pathogens has been somewhat of a controversial subject; more so among tree fruit pathologists in general than among those who have actively studied Cytospora canker. The opinions and attitudes of many pathologists were summarized 87 years ago in a statement made in assessing a New York fruit disease survey (Stewart *et al.* 1900). "Dead peach branches are commonly infested by a species of *Cytospora* which is generally believed to be a saprophyte, and considered of little importance; but we have so often seen this fungus intimately associated with dead and dying peach trees when no other sufficient cause for disease was evident, that we are becoming suspicious that it is, at least, a semiparasite".

Research for the past 87 years has clarified the role of Cytospora canker as a very destructive disease of stone fruit trees. It seems to be

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generally agreed that Cytospora canker development requires a stressed or weakened tree. However, once established in such a tree Cytospora canker is often the cause of death. The exact degree of stress or weakening required to predispose a tree to death and destruction from Cytospora canker is controversial and generally not known. Among the factors known to predispose stone fruit trees to Cytospora canker are winter injury, drought stress, nematodes, nutrient imbalances and possibly even cropping (Bertrand and English 1976a, Bertrand *et al.* 1976, English *et al.* 1982, Hildebrand 1947, Rolfs 1910, Wisniewski and Wilson 1985). It is of interest to note in regard to the latter as a possible stress mechanism that in the central valley of California, where cold injury does not occur, European plums (cv. 'French') almost never show Cytospora canker until they begin bearing.

There is a critical difference between a factor that predisposes a tree to aggressive invasion by Cytospora canker and a localized condition that provides a site for infection. Known sites of infection include pruning cuts, leafscars, insect injuries, sunburns, winter injuries, mechanical injuries, and dead twigs (DeVay *et al.* 1974, Hildebrand 1947, Rolfs 1910, Travis and Hickey 1985a, Tekauz and Patrick 1984, Willison 1937). Probably any wound could provide an infection court for *L. persoonii* or *L. cincta*.

Infection as defined by the landing and germination of a spore in a wound is a random event. Spores blown about in wind or splashed about in water are just as likely to land one place as another. The factors that determine whether an infection will sooner or later heal over or become an aggressive killing canker are temperature and the nebulous undefinable morass of "tree vigor" or "tree health". Several studies have addressed this issue. A California study (Bertrand and English 1976a) of Cytospora canker in French prune suggested tree health factors were important. In September 1971, 76 isolates of *L. persoonii* were inoculated into trees in an orchard free of Cytospora canker. The inoculation resulted in various sized cankers up to 7.0 cm long by the following January. By June 1972, all cankers had fully healed at the margins and none of them became active again. In a later study an isolate of *L. persoonii* selected during the previous work was inoculated into limbs of French prune trees in an orchard where Cytospora canker was a common problem and an orchard Cytospora canker was a non-problem. These inoculations were made March, 1973, and evaluated June, 1973. In the orchard where Cytospora canker was not a problem the cankers resulting from inoculation reached 5.6 cm in length before completely healing around the margins. The inoculations in the orchard where Cytospora canker was a problem resulted in cankers 10.7 cm long which showed no sign of healing. The continued activity of these cankers resulted in the death of major portions of inoculated trees. The first group of trees had enough vigor or health to heal out the cankers. The second group did not. The reason why, in terms of tree physiology, is the mystery. Some factors that predispose trees to Cytospora canker have been documented. The effect these factors have on tree health has not been defined.

Two studies on Cytospora canker in peach gave conflicting interpretations as to the factors that regulate the seasonal activity of Cytospora canker in peach. Jones and Luepschen (1971) concluded that seasonal activity of Cytospora canker in peach is regulated by growth, presumably healing, activities of the host. Wensley (1964), on the other hand, concluded that temperature was the primary regulating factor. These two studies taken with the results of the California study (Bertrand and English 1976a) could lead one to conclude, with some logic, that both factors operate more or less independently to regulate the seasonal activity of Cytospora canker. Temperature has a year round effect on the growth rate of Cytospora cankers. Cankers are more active when temperatures are near optimum for fungus growth. In trees with enough vigor to heal around a canker, whether or not the healing is enough to terminate canker activity, healing reactions retard canker activity during the periods of greatest cambial activity (Bertrand and English 1976a, Northover 1976, Wensley 1965). In a healthy tree, the healing reactions that restrict canker enlargement can act in opposition to and overcome the effect of temperature in promotion of fungus growth. On the other hand in trees with no capacity to heal around a canker temperature alone is probably the major determinant of seasonal canker activity (Bertrand and English 1976a, Wensley 1964).

Control of Cytospora canker has proven very difficult. In general chemical treatments have not been particularly successful (Hickey and Travis 1985, Northover 1976, Travis and Hickey 1985b). This may be due in part to lack of a specific time of infection. Wounds occur throughout the year from various causes (Hildebrand 1947, Willison 1937), infectious spores are released whenever rain occurs (Bertrand and English 1976b), and these spores germinate over a wide range of temperature (Hildebrand 1947). Surgical canker removal has been successful (Hildebrand 1947, Travis and Hickey 1985b) but surgically cleaned cankers would be as open to reinfection as any wound. Surgical canker removal has questionable practical value in a large orchard situation. Most Georgia pruning crews cannot distinguish dead from living limbs in the absence of leaves. Other means of controlling Cytospora canker such as "maintain good tree vigor" are certainly easier said than done, especially where winter injury is a chronic, nearly annual, problem. To achieve control of Cytospora canker, the major predisposing factors have to be dealt with to some extent on a local or regional basis (Bertrand *et al.* 1976, DeVay *et al.* 1974, Hildebrand 1947, Rolfs 1910, Rosenberger 1982, Travis and Hickey 1985a, Willison 1937).

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VARIATION IN VIRULENCE OF LEUCOSTOMA SPECIES.

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INTRODUCTION

Cytospora canker of peach, caused by Leucostoma cincta or Leucostoma persoonii, is the most destructive disease of peach in Michigan orchards. The disease is characterized by extensive perennial cankers on the trunk and branches, branch dieback, and progressive weakening of the tree. Fungicides have had no effect on the incidence of new infections or on the expansion of established cankers.

Biological controls have been gaining wider appreciation and acceptance (Cook and Baker 1983). For instance, natural biological control of another devastating canker disease, chestnut blight, appears to be responsible for the survival of the blight-susceptible European and American chestnut trees (Anagnostakis 1982). Less virulent or hypovirulent strains of the blight fungus, Endothia parasitica can be found on the trees within these recovering stands (Fulbright et al 1983). Hypovirulence has been defined as some measurable loss of virulence in a pathogen due to the presence of transmissible, cytoplasmically-borne, double-stranded RNA molecules (Anagnostakis 1982, Elliston 1981).

A method of control for Cytospora canker of peach might be through the use of hypovirulent strains of Leucostoma. The goal of our present research has been to identify a hypovirulent strain of Leucostoma and utilize it as a biological control of the cytospora canker disease. As in chestnut blight on chestnut trees, such strains could alter the disease severity and allow the natural defense mechanisms of the peach tree to ward off the pathogen.

MATERIALS AND METHODS

Isolates of Leucostoma spp. from peach trees were collected and rated for virulence on established trees, excised twigs, and apple fruit.

On November 8, 1985, ten Garnet Beauty peach trees were each inoculated with 24 different Leucostoma strains. Each inoculation was made on a separate branch and each branch measured 17 mm in diameter. Trees were wounded to xylem depth with an empty hand-held stapling gun. The wound area was frozen with a commercial aerosol freezing product (Scorza and Pusey 1984). Wounds were inoculated with a 5-mm mycelial plug and wrapped in Parafilm to prevent desiccation. On May 21, 1986, all of the inoculated branches were measured for canker length (length of necrotic area distal to inoculation point).

The peach twig virulence test was performed by inoculating stem pieces aprox. 12 cm long X 1-2 cm in diameter with mycelium plugs embedded in agar. The area to be inoculated was swabbed with 95% ethanol and flamed briefly. Using a 4mm cork borer a wound was made to xylem depth and the mycelial plug was placed in the wound and covered with masking tape. The twigs were placed in an open plastic bag for 2-3 weeks and the resulting lesions were measured.

The apple fruit virulence test was performed by removing a 9mm diameter X 7mm deep plug of `Golden Delicious' apple tissue and placing mycelia embedded in agar plugs in the wound (Fulbright 1984). The wound was taped to prevent desiccation. After the inoculated apples were placed in an open plastic bag and allowed to incubate at 25 C for 10 days, the width of the resulting lesions were measured.

Some isolates were additionally screened for the presence of dsRNA. Extraction of the dsRNA was accomplished by modifications of the double-cellulose column procedure (Dodds 1970, Fulbright et al 1983). The dsRNA was layered on Polyacrylamide slab gels (5%) and electrophoresed for 12 hr at 40 mA.

RESULTS AND DISCUSSION

Isolates of Leucostoma spp. from a number of locations in Michigan, North Carolina, West Virginia and California were ranked in order of virulence in orchard tree, excised twig and apple fruit virulence assays. Significant differences among the isolates were observed in the various tests. The twig and apple fruit tests yielded the most consistant order for ranking of virulence (Tables 1 and 2). Isolate 14.1 was ranked as most virulent, isolates 8.2 and 9.2 were moderately virulent and isolate 14.4A was least virulent in each test. Isolate 9.2 was more virulent in the apple fruit assay than in the twig assay; however, it demonstrated moderate virulence in both tests. The excised-twig test failed to statistically differentiate virulence in all but one isolate, 14.4A, which was statistically less virulent than the other isolates tested.

In orchard tree virulence assays trees of different germplasms varied significantly in susceptibility (P =.01) (Chang et al 1986). Trees of a common germplasm also varied significantly in susceptibility (P =.01), while branches within a single tree did not. Different isolates varied significantly in virulence (P = .01). These assays also allowed ranking of the isolates. Many of these rankings, however, conflicted with the twig and apple fruit assays (Table 3). For example, isolates 8.2 and Riley were statistically more virulent than other isolates in the orchard tests, but in the apple tests these isolates showed moderate virulence.

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Isolate 9.2 was rated as low in virulence in the orchard test. Also, isolate 14.4A which was statistically less virulent than all the other isolates in the apple fruit and twig tests was not statistically different in virulence than isolates 14.1, and 9.2 in the orchard test.

While each assay may provide a different virulence ranking of many of the isolates, one isolate, 14.4A was least virulent in all three tests. Isolate 14.4A is a nonsporulating fungus isolated from the bark of a scale-injured North Carolina peach tree by Endert-Kirkpatrick and Ritchie. The isolate (14.4A) was tentatively identified as Leucostoma cincta based on cultural morphology (Willison 1936). This isolate has a degenerative disease characterized by abnormal growth rate, swollen and often lysing hyphae,

Table 1. Mean comparison of lesion diameters in apple fruit between strain 14.4a and normal, dsRNA-free strains[a].

Strain	Lesion Width[b]
14.1	8.33 A[c]
Tubb's	7.77 AB
9.2	6.80 BC
10.8	6.67 CD
11.7	6.58 CD
Riley	6.53 CD
8.2	6.48 CD
Redhaven	6.08 CD
Mears	5.72 D
14.4A	1.52 E

a/ comparisons were made 10 days from inoculation
b/ means based on ten replicatons
c/ means followed by same letters are not significantly different (P=.01)

Table 2. Mean comparison of canker length in excised peach wood of strain 14.4A and normal dsRNA free strains[a]

Strain	Canker length[b]
14.1	7.35 A[c]
10.4	7.30 A
10.2	7.17 A
8.2	7.00 A
9.2	5.82 A
4A	5.17 A
14.4A	1.18 B

a/ comparisons were made 21 days from inoculation
b/ length of necrotic area distal to inoculation; means of seven replications
c/ means followed by same letters are not significantly different (P=.01)

Table 3. Mean comparison of canker lengths after inoculation of 10 Garnet Beauty peach trees with twenty-four Leucostoma isolates.

Strain	Canker length[a]	DsRNA[b]
Riley	11.00 A	-
8.2	9.98 AB	-
F-46	9.83 ABC	-
Class	8.40 BCD	-
10.8	7.78 BCDE	-
H9.5	7.64 BCDEF	-
C-J-1	7.50 CDEF	-
C-S-20	7.24 DEFG	-
Gum	7.08 DEFGH	-
Fenn	6.59 DEFGHI	-
Ma-4	6.49 DEFGHIJ	-
C-MI-5	6.48 DEFGHIJ	ND
PA	6.06 DEFGHIJK	-
C-jm-18	5.94 EFGHIJK	ND
H10.9	5.46 EFGHIJK	ND
9.2	5.34 FGHIJK	+
14.1	5.11 GHIJK	-
F-45	4.92 GHIJK	-
14.4A	4.81 HIJK	+
H7.13	4.79 HIJK	-
H6.15	4.75 HIJK	-
H9.11	4.71 IJK	-
11.11	4.18 JK	-
I-80	4.02 K	-

a/means followed by the same letters are not significantly different (P=.01)
b/-,no dsRNA present; +,dsRNA present; ND, not determined

reduced or absent conidiomata, and reduced virulence. This isolate also contains dsRNA as determined by gel electrophoresis (Figure 1). The hyphae of strain 14.4A contains spherical virus-like particles at high densities, as observed in ultra-thin sections with the electron microscope. The virus-like particles are 32.0 ± 3 nm in diameter, frequently exhibit an electron dense core and are surrounded by a capsid (Snyder et al 1986).

Attempts to associate the dsRNA present in 14.4A with the abnormal culture morphology have been complicated by the number of dsRNA molecules found in this strain. It is possible to eliminate both dsRNA and the hypovirulence phenotype in E. parasitica by single-conidium isolation or treatment with cycloheximide (Fulbright 1984). However, with isolate 14.4A this has proven difficult since the isolate does not produce spores and is highly sensitive to cycloheximide. Instead, hyphal tipping of 14.4A resulted in fewer dsRNA bands and decreased dsRNA concentrations. The spherical virus-like particles that filled the hyphae of the original 14.4A isolate were no longer visible through electron microscopy in the hyphal-tipped cultures. The partially cured isolate showed increased virulence in excised peach wood and apple fruit. The hyphal-tipped isolate appears morphologically normal and readily forms pycnidial structures; however, no conidia have been produced.

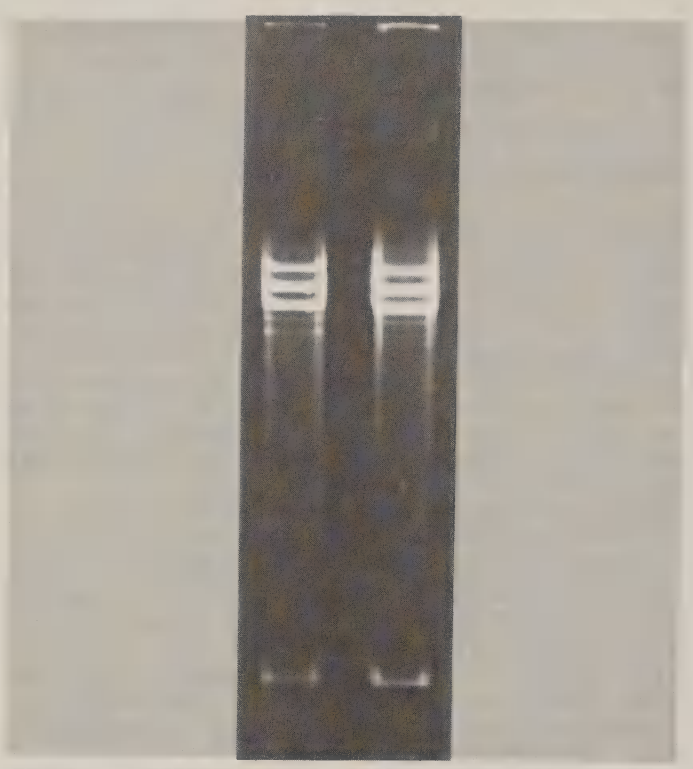

Figure 1. DsRNA banding pattern of isolate 14.4A.

Selection of colonies derived from regenerated protoplasts of 14.4A yielded two virulent sporulating, morphologically normal isolates. One strain has no detectable levels of dsRNA as analyzed by gel electrophoresis. The other strain has fewer bands than the original 14.4A and they appear much fainter in the polyacrylamide gel. This suggests a decrease in the dsRNA concentration. Initial virulence tests in apple fruit indicate that these two cultures are more virulent than the original 14.4A isolate and its derived hyphal tipped isolate. Comparing the culture morphology and virulence of the original 14.4A with the morphologies and virulence of the new isolates with various molecules of dsRNA eliminated should indicate which dsRNA molecules are responsible for the changes observed.

The presence of dsRNA alone is not enough to cause changes in virulence and morphology in Leucostoma spp. Isolate 9.2 contains dsRNA and has normal morphology, sporulates readily, and exhibits normal virulence. It is the only other isolate found to contain dsRNA in this study (Table 3).

Although isolate 14.4A is less virulent than normal isolates of Leucostoma spp. tested, it should not at this time be considered hypovirulent since the hypovirulent phenotype has not been transferred to other strains. This work is in progress.

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245 AMBROSIA BEETLES: PROBLEM OR SYMPTOM?

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INTRODUCTION

More than 6,000 species of scolytid beetles (including "bark beetles" and "ambrosia beetles") have been described and about 480 species occur in the United States (Wood 1982). Approximately 70 species occur in South Carolina, but only two bark beetles, the shot hole borer (Scolytus rugulosus {Muller}) and the peach bark beetle (Phloeotribus liminaris {Harris}) are considered to be pests of peach trees (Kirk 1969, 1970).

Bark beetles differ from ambrosia beetles mainly by their feeding habits. Bark beetles generally burrow and feed in the phloem tissue of the inner bark. Eggs are laid in clusters and larvae bore tunnels at right angles to the egg gallery as they feed on the phloem. The adult beetles exit the tree from separate round holes. After the brood emerges the surface of the infested tree appears peppered with shot holes (Wood 1982).

Ambrosia beetles tunnel directly into the sapwood and feed primarily on "ambrosia" fungi that line the wall of their tunnels. The ambrosia beetles have specialized structures (mycetangia) in which spores of the fungus are carried. As the female bores into the wood, usually at a lenticel in peach trees, fungal spores are deposited on the walls of the tunnel.

The female continuously grazes the fungal mat to prevent the fungus from filling the tunnel system and suffocating her progeny. The larvae wander inside the tunnels and form pupal cells along the gallery walls. If mating occurs, it must take place before the young females leave the gallery system since males are unable to fly and usually die within the trees. A generation is completed in 30 to 40 days under favorable conditions. Scolytids normally attack damaged or unhealthy trees, but some species attack healthy wood and may be of economic importance (Fischer 1954, Francke-Grosmann 1967, Wu et al. 1978, Wood 1982).

Ambrosia beetles were first identified as attacking peach trees in March 1982 in Florence County, South Carolina. Numerous second leaf trees were in a weak or dying state with sawdust-like frass strands protruding 2 to 4 cm from small holes in the lenticels. Some trees were dead from the ground up, while others were putting out new leaves and producing large amounts of gum from the entry holes. In most cases, the gumming trees overcame the beetle attack and survived. Extensive surveys indicated that infestation of peach trees was quite widespread throughout the state, involved several species of scolytid beetles, and that infestations were found in orchards with obvious symptoms of stress or decline as well as apparently healthy orchards.

Concerns were raised by research and Extension personnel, as well as peach producers, as to the potential for scolytid beetles becoming serious pests of peaches. Studies were undertaken to determine the species composition of scolytid beetles attacking peach trees, their biology, distribution and effect on peach trees in South Carolina.

MATERIALS AND METHODS

Vaned window traps (Kovach 1986) baited with 75% ethanol were used to survey scolytids occurring in South Carolina peach orchards. Traps were located in orchards in each of the three primary peach producing regions of the state (Kovach 1986) .

To provide data on the number of species actually infesting peach trees, infested wood was collected and returned to the laboratory, where it was either dissected or held in a rearing room for adult emergence. Polystyrene vials with 1-cm openings cut in the caps were placed over individual holes to act as emergence cages since ambrosia beetle progeny exit the tree via parental entrance holes.

In peach orchards containing infested trees, general observations on tree location, orchard sanitation, orchard drainage and cold injury were recorded and soil samples taken to assess soil pH and nematode populations. These observations were used to determine potential stress factors associated with infested trees.

To attempt to assess the physical damage associated with ambrosia beetle tunnelling and its affect on peach trees, 2-mm holes were drilled to a depth of 2 - 3 cm in the trunks of second leaf 'Blake' peach trees. Hole densities of 50 and 100 per tree were used. Holes were drilled in the fall or spring.

Three growth parameters were measured throughout the growing season; primary growth or shoot length, secondary growth or trunk circumference and flower bud set. Primary growth was measured from the same three shoots/tree by counting the number of leaves per shoot (Haun and Coston 1982). Secondary growth was measured as trunk circumference 15 cm above the soil, and flower set was measured as the number of flower buds per cm of shoot.

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RESULTS AND DISCUSSION

Twenty species of scolytids were found in South Carolina peach orchards. They were Phloiotribus liminaris (Harris), Scolytus rugulosus (Muller), Monarthrum fasciatum (Say), Monarthrum mali (Fitch), Xylosandrus crassiusculus (Motschulsky), Xylosandrus germanus (Blandford), Xylosandrus compactus (Eichhoff), Hypothenemus dissimilis (Zimmermann), Xyleborus ferrugineus (Fabricius), Xyleborus affinis Eichhoff, Xyleborus celus Eichhoff, Xyleborus dispar (Fabricius), Ambrosiodmus tachygraphus (Zimmermann), Ambrosiodmus rubricollis (Blandford), Xyleborinus saxeseni (Ratzeburg), Pseudopityophthorus asperulus (LeConte), Cnesius strigicollis LeConte, two Hypothenemus spp. and Pityophthorus sp.

The bark beetles, S. rugulosus and P. liminaris, and the ambrosia beetles, Xyleborinus saxeseni, Xylosandrus crassiusculus, A. rubricollis, A. tachygraphus, Xyleborus dispar, and M. fasciatum were found to infest peach wood. The four most common ambrosia beetles were X. saxeseni (90.4% of the total), X. crassiusculus (4.9 %), M. fasciatum (4.3 %) and A. tachygraphus (0.4%). A key to the common species is found in Kovach and Gorsuch 1985.

Xyleborinus saxeseni was more common in the Piedmont, 72% of the trapped population. Seventy-three percent of A. tachygraphus were trapped in the ridge counties while 89% of X. crassiusculus were from the coastal plain counties. Populations of M. fasciatum were evenly distributed throughout the state.

Xyleborinus saxeseni is a native of Europe and has apparently been present in South Carolina since the early 1900's. It is found in most areas of the U.S. and southern Canada. It has 4 or 5 generations per year in South Carolina and attacks weakened peach wood that average 4.8 cm diameter. Initial emergence occurs when maximum temperatures for two consecutive days are greater than 21° C (usually in early March). Subsequent emergence peaks occur every 57 days through November. The host range includes most deciduous as well as pine trees.

Xylosandrus crassiusculus is a recent introduction from east Africa and southern Asia. It was first reported in the continental U.S. from Charleston and Dorchester Counties, South Carolina,in 1974 (Anderson 1974). This beetle is capable of attacking young, apparently healthy peach trees. It is univoltine with peak emergence in early March and appears to undergo an obligatory diapause in South Carolina. It has also been reported from cherry and sweetgum in South Carolina.

Both A. tachygraphus and M. fasciatum are native species found throughout much of the eastern U.S. Both have relatively wide host ranges.

Most trees infested with native or long established ambrosia beetles had some other stress factor(s) associated with them. Factors included cold injury, ring nematode, Criconemella xenoplax (Raski) Luc and Raski, populations ranging from 250 to 2520 per 100 cc of soil, acidic soil (pH from 4.8 to 5.6) and low nutrient levels in the soil. No stress factors could be associated with trees infested with X. crassiusculus or A. rubricollis.

Simulation of mechanical injury associated with ambrosia beetle tunnelling by drilling holes in second leaf peach trees showed the following. Holes drilled in the fall had no influence on the primary growth of peach trees the following season compared to control trees. Drilling 50 holes in the spring increased vegetative growth 28% over the growing season.

Secondary growth was increased by as much as 44% compared to control trees regardless of the timing or density of holes drilled. The largest increase in trunk circumference (6.57 cm) occurred in trees that were drilled with 50 holes in the spring. Flower set was not reduced by drilling holes.

Relatively healthy peach trees often overcome attack by ambrosia beetles through the production of sap. The resulting gum often fills the tunnel system and drowns the beetles. New growth will cover the entry hole. However, these trees will ooze balls of gummy sap from the lenticels creating a situation that is easily mistaken for fungal gummosis. To differentiate the two causes it is necessary that thin serial cuts be made through the bark and into the woody tissue.

If fungal gummosis is the cause, the gummy pocket will not extend into the woody tissue. If ambrosia beetles are the cause, a thin, vertical stain will be found in the woody tissue. It may be necessary to cut completely through the previous season's growth before the tunnel is found. The tunnel will be completely filled with sap.

From our work, it appears that most attacks by ambrosia beetles are symptoms of other problems in a peach orchard. The primary exception is if the attack is by Xylosandrus crassiusculus. This ambrosia beetle appears to be quite aggressive and capable of attacking relatively healthy trees. Under proper environmental conditions, the trees are not able to overcome the attack and succumb to the beetles.

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THE PHEROMONE PERMEATION TECHNIQUE AS A CONTROL FOR THE PEACH TREE BORER[1/]

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ABSTRACT

In 1984 and 1985, experiments were conducted in peach orchards to control the peachtree borer (PTB), Synanthedon exitiosa (Say), with its pheromone, (Z,Z)-3,13-octadecadienyl acetate (Z,Z-ODDA). The pheromone was dispensed in 2.54 cm^2 vinyl laminated dispensers manufactured by the Hercon Corporation (Gentry et al. 1980, Snow et al. 1985). An average evaporation rate of 0.29 mg/day/dispenser was obtained or ca 104 mg/ha/day. During these experiments, ca 75 percent control of the species was obtained when all the infested orchards in an area were treated. The treatment of wild hosts adjacent to the orchards was not necessary to obtain control. This new method of control offers a non-insecticide method to control PTB that is estimated as price competitive with insecticides.

INTRODUCTION

Research in the use of atmospheric permeation with pheromone to disrupt mating of the PTB was initiated about 10 years ago. Excellent results have been reported by McLaughlin (1979), McLaughlin et al. (1976), Gentry et al. (1980), and Yonce (1981), when control was based on reduction of numbers of males captured, or when treatment was initiated in young orchards before they became infested. However, in all these cases, economic control was not recorded or did not occur over long periods of time. The tests reported here were conducted to determine if economic control of the PTB could be obtained when entire orchards and wild host areas around the orchards were treated with pheromone.

MATERIALS AND METHODS

In all our experiments, Z,Z-(ODDA) was formulated commercially and loaded (ca 43 mg/dispenser) into 2.54-cm^2 vinyl laminated dispensers. Experiments were conducted in three different blocks of peach trees located in central Georgia, and each tree (in 2 blocks) was treated with one dispenser per tree. In one treatment area the wild hosts (plums and black cherries) within 1.5 km of the area were treated but were not in the other treatment area. This scheme was reversed in the second year. Male PTB populations were monitored using Pherocon 1C sticky traps baited with the same pheromone. A single control orchard was maintained for the two treatment orchards.

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In 1984, mating activity was determined by using mating tables (Snow et al. 1976, Snow et al. 1985). Tables were operated both within the orchards and outside the orchards (within the 1.5-km-radius area) in wild host areas. Female PTB were collected from orchards with insect nets. Where appropriate, a two-way analysis of variance ($P<0.05$) or X^2 contingency table analysis ($P<0.01$) was used to analyze the data.

Area M. This treatment area consisted of 6 orchards comprising 44.8 ha. that were moderately infested with PTB (reason called Area M). We treated the entire 44.8 ha in 1984 and in 1985 we treated the same 44.8 ha, but we also treated the wild host areas within 1.5 km of the central orchard. The nearest untreated peach orchard was 20 ha in size and located about 6.0 km away.

Area H. This treatment area consisted of seven orchards, but all were young and lightly or non-infested except for a 19.2-ha orchard that was heavily infested with PTB (the reason called Area H). During 1984, we treated the entire area including the wild host areas within a 1.5-km radius area, but in 1985 the wild host areas were excluded.

RESULTS

The release rate of pheromone is shown in Figure 1 as mg retained and released per 2-week period (average of 3 samples per time period). During this entire placement time in the field, the evaporation rate averaged 0.29 mg/day or ca 78.0 mg/ha/day (31.6 mg/acre/day). The release rate was basically linear during the entire testing period. No males were captured in either treatment orchard after placement of the pheromone between August 1-10 (Fig. 2).

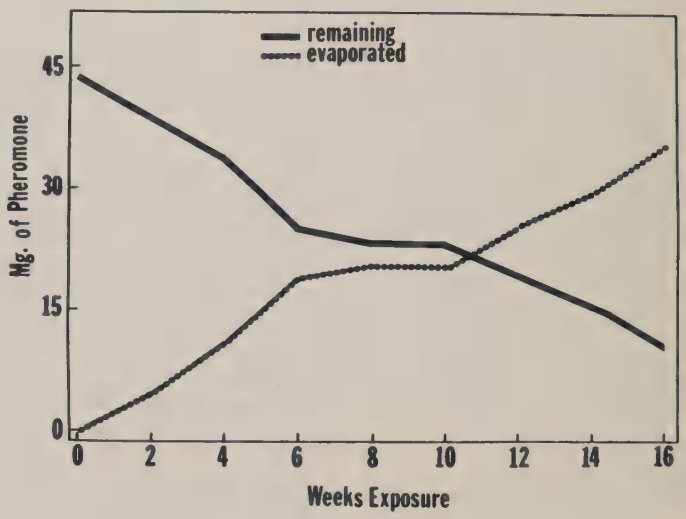

Figure 1. The retention and evaporation of the pheromone, (Z,Z)-3,13-octadecadienyl acetate, per dispenser, hung in a peach orchard during 1985 (0 weeks = 43.4 mg).

Data collected from mating tables is shown in Table 1. More females were placed in some locations than in others because data from these locations were more critical and because the number of available females was limited. In area M, the moderately infested orchard where the adjacent wild hosts were not treated, no mating occurred in the orchard,but 23% of the females mated in the wild host area within the 1.5-km-radius area. In Area H, the heavily infested orchard, 2% of the exposed females mated in the orchard but none mated on the tables in the wild host area (which were treated) within the 1.5-km-radius area. Females on mating tables in the control orchard were 84 percent mated.

Figure 3 shows the data when females were collected with sweepnets. In 1984, 70 females were collected in the control orchard and 8.6% of these were virgins. In Area M, 70% of all collected females (28 of 40) were virgins, and, in Area H, 52% (69 of 132) were virgins. In 1985, in Area M (which in this year had the wild hosts treated) 76% percent of the collected females (45 of 59) were virgins; and in Area H where the wild hosts were not treated, 79% (31 of 39) of the collected females were virgins. The control had 12% virgins (19 of 160). In general, the populations were much lower in both orchards during 1985 and the collection of adults was very difficult.

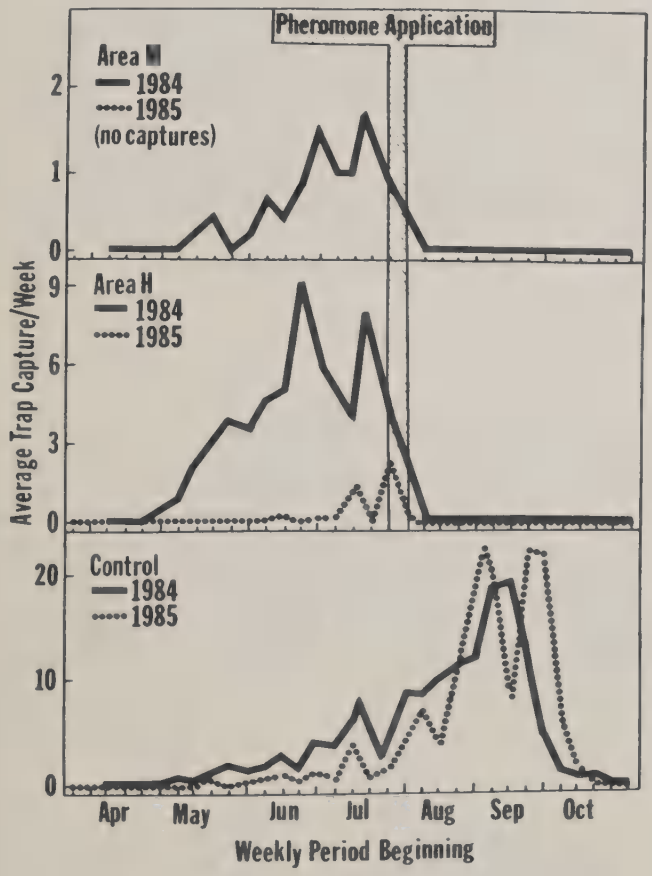

Figure 2. The capture of male PTB in monitor traps in the 3 test areas, 1984 and 1985.

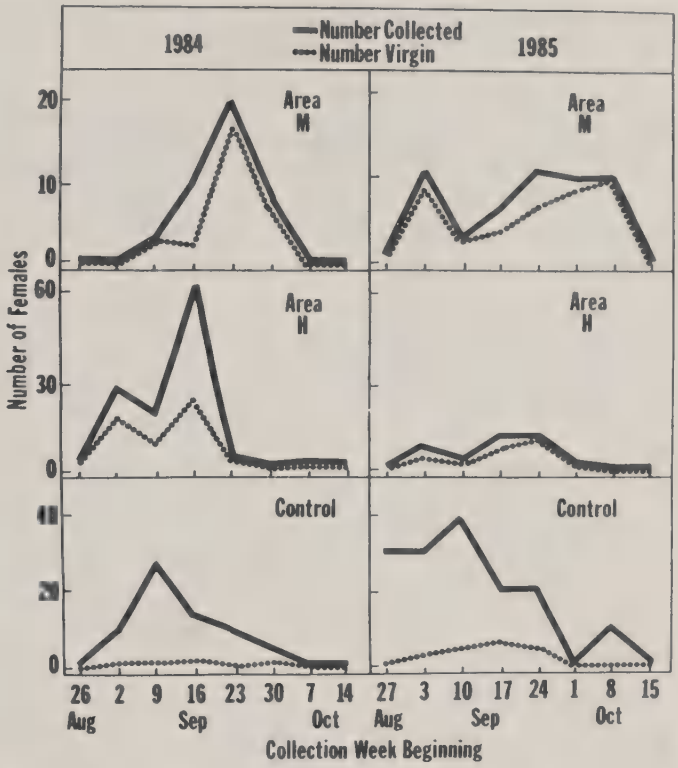

Figure 3. The collection by date and mating status of wild females collected with sweepnets from the treatment areas in 1984 and 1985.

Control was almost equal both years in Area M regardless of whether the wild host area was treated or not. In neither year was mating observed in this area on mating tables (1984) or by observations (1984-85). However, in Area H the control level varied (52 and 79%) during the 2 years. While the reduction in mating during 1984 is low, it is more significant than first perceived because of the many mortality factors that come into play against eggs, larvae, and pupae. The good level of control and difficulty in finding adults the following year (1985) are evidence of the effect that the 1984 treatment had in Area H. These data indicate that effective control of PTB was achieved in both treatment areas regardless of whether the native plums and wild cherries around the orchards were treated. I interpret this to mean that in most situations, pheromones are a valid control procedure and that wild hosts do not contribute enough insects to overcome this control. However, I do believe that all the infested orchards within an area must be treated to result in good control (if they contribute significant numbers of mated females). Sufficient movement between adjacent orchards has been established to offset the effects of control if they are left untreated.

Table I. Collection of mated pairs of PTB from mating tables operated in and around test areas, 1984.

Mating Table Location	Days Data Collected[1/]	Females[2/] Unmated	Mated	% Mated	Separation of Counts by X^2
		Area M			
Within	10	100	0	0	a
Outside	8	60	18	23	b
		Area H			
Within	10	100	2	2	a
Outside	8	43	0	0	a
		Control			
Within	9	8	42	84	c

1/ A day's data is considered one table per treatment with 5 to 10 wing-clipped females.
2/ Accumulated totals in each category. Totals are significant at ($P<0.01$).

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Technique

ELECTROPHORETIC ANALYSIS OF CRUDE PROTEIN EXTRACTS FOR THE IDENTIFICATION OF PEACH ROOTSTOCKS.

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The use of 'Lovell' rootstock is one of several cultural practices recommended for managing peach tree short life (PTSL) in South Carolina and throughout most of the southeastern states (Brittain and Miller, 1976, 1978, Spivey and McGlohon 1973). This recommendation is made due to Lovell's superior performance on PTSL sites in comparison to other peach rootstocks including 'Nemaguard'. Therefore, accurate identification of peach rootstocks is of significance to growers for the effective management of this tree decline problem. At present, peach growers must rely on the supplying nursery to ensure that rootstock requests are correctly provided.

Electrophoretic analysis of plant proteins has previously been utilized to identify species and cultivars of petunia (Natarella and Sink 1975), cultivars of strawberry (Bringhurst et al. 1981), scion cultivars (Weeden and Lamb 1985) and rootstocks of apple (Weller and Costante 1986), and scion cultivars of peach (Carter and Brock 1980). The objective of this study was to determine the suitability of isoelectric focusing (IEF), discontinuous (DISC) gel electrophoresis and SDS-polyacrylamide gel electrophoresis (SDS-PAGE) for the identification of 5 rootstocks using crude protein extracts from peach bark and wood tissue.

MATERIALS AND METHODS

The rootstocks examined in this study were 'Nemaguard', 'Lovell', 'Tennesee Natural', 'Siberian C' and 'Boone County'. Plant material was collected from the seed certification and germplasm blocks located at the Sandhill Research and Education Center near Columbia, South Carolina. All samples were obtained on April 11, 1986. Ten 30.5-cm cuttings of one-year-old wood were collected from mature trees of each rootstock cultivar. Cuttings were immediately placed on ice and transported to Clemson University.

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Bark tissue and the outermost layers of wood were removed from each cutting with a budding knife and the remainder of the twig was discarded. The shavings were placed in glass jars and freeze dried for 3 days. The peach tissue was then ground into a dust and freeze dried again for a period of 2 days. After freeze drying the ground peach tissue was stored at -20 C and protein was extracted on a periodic basis from this material.

Extraction of peach proteins was conducted using a 0.1M Tris-HCl buffer with the addition of 0.7 g ascorbic acid, 0.1 g cysteine, 17.1 g sucrose and 0.05 g disodium ethylenediaminetetraacetate per 100 ml of buffer. The final buffer pH was adjusted to 8.3 prior to use. Protein was extracted from 500mg of peach tissue into 6 ml of extraction buffer. Peach tissue was homogenized with a Brinkman polytron and protein was extracted into the buffer for a period of 1 h at 4 C. Following protein extraction, the samples were centrifuged in a Sorvall RC2-B refrigerated centrifuge at 15,000 g for 30 min. The supernatant was collected and recentrifuged at 15,000 g for an additional 30 min. A 1-ml sample of the resulting supernatant was added to the sample resevoir of a Centricon-30 microconcentrator (Amicon) and centrifuged at 5,000 g for 25 min. That portion of the sample which was retained in the sample resevoir was utilized as the crude protein extract.

Discontinuous and SDS-PAGE was carried out on a Bio-Rad Protean II slab gel electrophoresis cell. Gels were 16 cm in length and consisted of a 1.5-cm 4% acrylamide stacking gel, a 2.0-cm 6.3% acrylamide spacer gel and a 12.5-cm 11% acrylamide resolving gel. All gels contained a cross-linkage of 4%. SDS polyacrylamide gels contained 1% SDS. A sample concentration of 100 micrograms protein was added to each sample well. For SDS-PAGE, 1% SDS and 5% beta-mercaptoethanol were added to each rootstock protein sample and samples were heated at 100 C prior to electrophoresis.

Isoelectric focusing was conducted on an LKB 2117 Multiphor II electrophoresis unit. IEF gels contained 6% acrylamide with a cross-linkage of 3%, and a pH gradient of 2.6-5.8. Isoelectric focusing was conducted at a constant power of 25 watts for each gel. Electrode buffers were 0.1 M sulfuric acid and 0.1 M sodium hydroxide. IEF gels were prefocused for 1/2 h at which time sample applicators consisting of 7 X 20 mm strips of Whatman #3 filter paper were placed on the gel at the cathode end. A 100-microgram protein sample was applied by pipet to the individual applicators. Filter paper was removed from the gel after 1 h and gels were focused for an additional 2 h.

Protein bands were visualized using silver staining techniques. IEF and DISC gels were stained using the method of Nielson and Brown (1984) with the exception of the fixative solution. A 3.5% sulfosalicylic acid, 11% trichloroacetic acid solution was utilized as

the fixative. Gel staining was conducted at 40 C in a water bath. The method employed for staining SDS gels is found in table 1 and comprizes several steps from the procedure of Morrisey (1981) and that of Nielson and Brown (1984).

Table 1. Staining procedure for the visualization of protein bands in SDS-polyacrylamide gels.

1. 30 min rinse in 50% methanol, 10% acetic acid solution
2. 30 min rinse in 5% methanol, 7% acetic acid solution
3. Overnight soak in 10% glutaraldehyde
4. 2 h rinse in distilled deionized water
5. 40 min soak in 0.03 mM dithiothreitol
6. 30 min soak in 12 mM silver nitrate
7. Two 1 min rinses in distilled deionized water
8. Soak in 0.28 M sodium carbonate, 0.0185% formaldehyde
9. Soak in 5% acetic acid to halt development

RESULTS AND DISCUSSION

Protein banding patterns obtained from the separation of peach proteins by discontinuous gel electrophoresis and SDS-polyacrylamide gel electrophoresis were not sufficiently different between samples to obtain accurate identification of rootstocks. Differences could not be consistently detected either visually or through the analysis of tracings generated from the scanning of gels with a LKB Ultrascan XL laser densitometer. All bands were present for each rootstock,and although staining intensities of particular protein bands sometimes varied, the inconsistency of this characteristic prevented accurate identification of peach rootstocks.

Through the visual analysis of isoelectric focusing gels, the identity of individual rootstocks could consistently be determined based on protein banding patterns. All protein bands were located within the pH range of 3.2 to 4.3. The distinction of rootstocks could be made based on the relative position or presence of 4 protein bands, 2 at the cathode end and 2 at the anode end of the protein band area. 'Boone County' lacked a dark cathode terminal band and a dark anode terminal band present in the protein banding pattern of the other rootstocks. 'Tennessee Natural' lacked the dark cathode terminal band present in the protein banding pattern of 'Nemaguard', 'Lovell' and 'Siberian C'. 'Siberian C' contained 2 anode terminal protein bands while all other rootstocks contained only 1. 'Nemaguard' and 'Lovell' could be separated on the basis of relative position of the anode terminal protein band. The anode terminal protein band of 'Lovell' was found at a pI of approximately 3.4 while that of 'Nemaguard' had a pI of about 3.2.

In conclusion, the identity of peach rootstocks could not be determined based on protein banding patterns resulting from discontinuous or SDS gel electrophoresis. However, the identity of rootstocks could accurately be determined based on protein banding patterns resulting from isoelectric focusing of crude protein extracts from peach wood tissue. In the continuing search for resistant rootstocks, this technique may be a means to provide quality control for nursery operations should a superior rootstock be developed.

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